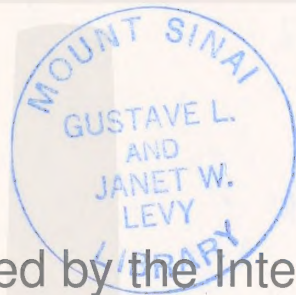


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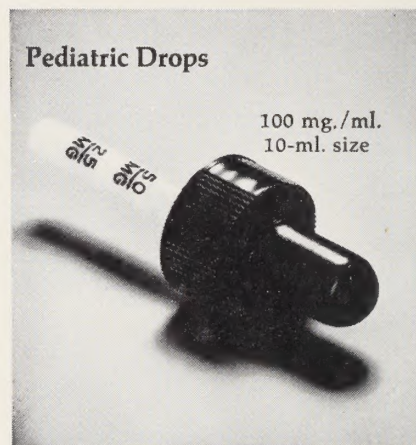
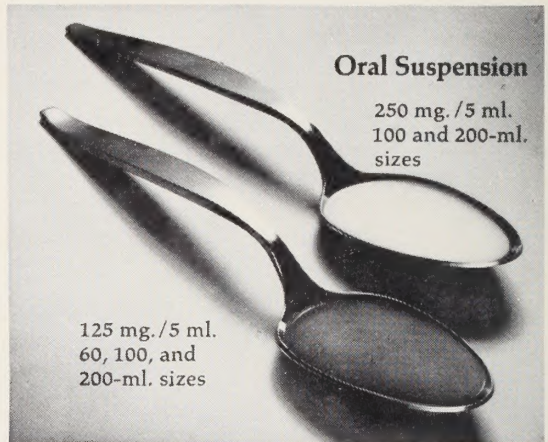
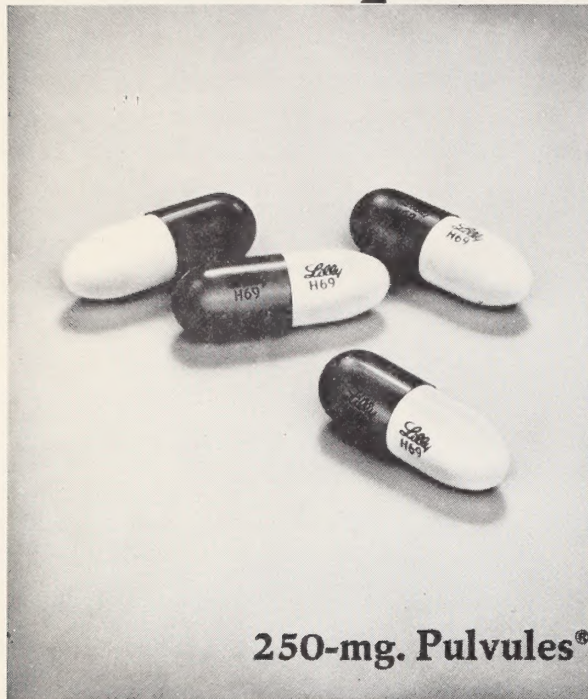
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An Assessment of Physician Knowledge of Tay Sachs Disease

ELLIOT D. ROSENSTEIN, M.D., LYNN GODMILOW, M.S.W., AND KURT HIRSCHHORN, M.D.

Abstract

A study was undertaken to evaluate the knowledge of Tay Sachs disease in the Mount Sinai School of Medicine medical community and the degree of physician participation in and advocacy of Tay Sachs screening. The results of a questionnaire distributed to 234 medical students, house staff and attending physicians were analyzed.

Physicians at Mount Sinai performed reasonably well, with pediatricians scoring substantially higher than the other specialists. The greatest deficiency of knowledge was noted in awareness of the carrier rate in the at risk population. This risk was significantly underestimated in our sample. This underestimation of risk is a possible explanation for physician's poor compliance with Tay Sachs screening when the physician himself was at risk as a carrier. This underestimation of risk is also offered as an explanation for the poor record of the physicians as advocates of screening for their at risk patients. In addition, and of particular interest, was the inadequate understanding of the mechanics of inheritance. A suggestion for the education of the medical community is offered.

Introduction

In his study of the utilization of the health system, Rosenstock observed that people use health services according to their perception of health (1). In the absence of clinical manifestations, a number of factors influence a person's decision to comply with a particular screening test: appreciation of the susceptibility to and severity of the condition, appreciation of the possible consequences of noncompliance, and appreciation of the potential benefits of participation in the program.

The physician has the potential for being the most powerful advocate of health programs. Compliance rates can be improved when advocates of the program are well informed and motivated. In an assessment of Tay Sachs disease screening in Montreal, Beck et al observed that the physician performed poorly in his role as advocate. Beck saw a need for improved perception of the personal relevance of heredity, not only for the patients of genetic screening programs, but also for the health care personnel who function as advocates (2).

It was our intent to determine the extent of

physician knowledge of Tay Sachs disease and to assess its implications for genetic screening programs. This investigation was particularly pertinent in view of Mount Sinai's long association with this disease and the substantial proportion of the Mount Sinai medical community who are members of the population at risk (3).

In 1887, Dr. Bernard Sachs, a neurologist at The Mount Sinai Hospital in New York, first presented a clinical report of a child with cherry red spots and progressive neurologic deterioration. Dr. Sachs characterized the disease, named it "amaurotic familial idiocy" and noted its preponderance among children of Jewish ancestry (4). Dr. Warren Tay, A British ophthalmologist, had described the cherry red spots earlier (5). Later, the disease came to be known as Tay Sachs disease.

This study was designed to answer the following questions: 1) Is there a relationship between the level of professional education or experience and the degree of knowledge about Tay Sachs disease? 2) Is there a relationship between areas of medical specialization and the degree of knowledge about Tay Sachs disease? 3) Are there specific aspects of Tay Sachs disease about which there are deficiencies of knowledge? 4) Are these physicians informed as to the mechanics of specific modes of inheritance? 5) Is there a relationship between specialty

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area and the decision to be tested? 6) Is there a relationship between extent of knowledge about Tay Sachs disease and the decision to be screened?

Materials and Methods

A single page questionnaire was prepared consisting of 17 objective questions; seven were to assess demographic data about the respondent, six were to ascertain knowledge of the respondent about the etiology, inheritance, ethnic proclivity, incidence and clinical manifestations of Tay Sachs disease, two questions were designed to assess the respondent's understanding of the mechanisms of inheritance and two questions were designed to elicit information about the respondent's participation in a screening program (appendix A).

The questionnaire was distributed to medical students, house staff and faculty at selected lectures and departmental conferences. Data from 234 respondents were collected; 66 medical students, 118 house officers and 50 members of the faculty. Ninety-five percent of those present at the conferences completed the form. Therefore, we feel there was no inherent bias in the selection process and that the sample questioned is representative of the medical community at Mount Sinai.

The questions assessing physician knowledge about Tay Sachs disease and modes of inheritance were scored correct only if the respondent chose the single most acceptable answer, or if he properly qualified his responses if more than one response was chosen. An answer was scored incorrect if more than one response was chosen without qualification, if a single wrong answer was chosen or if no response was chosen. One point was awarded for each correct answer. There was no partial credit and no penalty for a wrong answer. After the questionnaires were scored, the population was divided into subgroups by religious origin, hospital staff status and medical specialty. In addition, the Jewish population was further subdivided according to whether or not they had been screened. Statistical significance was determined by the *t* test.

Results

Overall performance on questions assessing knowledge about Tay Sachs disease is shown in Table I. The overall mean score of the population was 4.59 out of a total possible score of 6. The pediatric subgroup scored highest with a mean score of 5.71 and the psychiatric subgroup scored lowest with a mean score of 3.75.

Table II compares the performance of the physician subgroups on the knowledge of Tay Sachs

TABLE I
Overall Performance of Physician Population on Questions Assessing Knowledge of Tay Sachs Disease

Segment of Population	Number of Respondents	Mean Score $\pm$ S.D.
Jewish	172	4.69 $\pm$ 1.19
Screened	24	5.21 $\pm$ 0.72
Nonscreened	148	4.60 $\pm$ 1.23
Non-Jewish	62	4.32 $\pm$ 1.29
Attending Staff	50	4.74 $\pm$ 1.44
House Staff	118	4.54 $\pm$ 1.20
Medical Students	66	4.57 $\pm$ 1.10
Pediatrics	28	5.71 $\pm$ 0.53
Neurology	20	5.05 $\pm$ 0.94
Obstetrics/Gynecology	14	4.57 $\pm$ 1.16
Medicine	26	4.53 $\pm$ 1.03
Ophthalmology	22	4.50 $\pm$ 1.47
Surgery	42	4.05 $\pm$ 1.31
Psychiatry	16	3.75 $\pm$ 1.29
Total	234	4.59 $\pm$ 1.23

Maximum Score = 6.

TABLE II
Comparison of Physician Subgroups Performance on Questions Assessing Knowledge of Tay Sachs Disease

Segment of Population	Scored Better Than	No Significant Difference From
Jewish	Non-Jewish ($p < .025$)	—
Attending Staff	—	House Staff Medical Students
House Staff	—	Medical Students
Pediatrics	Neurology ($p < .005$) OB/GYN ($p < .005$) Medicine ($p < .005$) Ophthalmology ($p < .005$) Surgery ($p < .005$) Psychiatry ($p < .005$)	—
Neurology	Medicine ($p < .05$) Surgery ($p < .005$) Psychiatry ($p < .005$)	Obstetrics/ Gynecology Ophthalmology
Obstetrics/ Gynecology	Psychiatry ($p < .05$)	Medicine Ophthalmology Surgery
Medicine	Psychiatry ($p < .025$)	Ophthalmology Surgery
Ophthalmology	—	Surgery Psychiatry
Surgery	—	Psychiatry
Screened Jewish	Nonscreened Jewish— ($p < .01$)	—

disease. It was observed that the Jewish subgroup scored significantly better than the non-Jewish group ($p < .025$). No significant difference was noted in the hospital staff status subgroups. On the basis of medical specialty, pediatricians scored significantly higher than all other subspecialty groups ($p < .005$). In the Jewish subgroup, those who had been screened scored significantly higher than those who had not been screened ($p < .01$).

Table III reflects performance on the six specific questions pertaining to knowledge about Tay-Sachs disease. Of the six questions, 75% of respondents answered five correctly. The obvious exception was the question pertaining to the carrier rate which 64.5% of the population answered incorrectly (95% of whom underestimated the one in the thirty carrier rate).

Table IV demonstrates the performance of all respondents who knew the mode of inheritance on questions concerning the risk of carrier parents to have children affected with Tay Sachs disease. Of the 85% of respondents who knew that Tay Sachs disease was inherited in an autosomal recessive

pattern, less than 80% knew the risk of having an affected child if both parents were carriers or if only one parent was a carrier.

Table V illustrates the participation of the population surveyed in Tay Sachs screening. The overall compliance rate was 13.9%. The only subgroup with a significantly greater compliance rate (30.4%) was the pediatric group ($p < .05$).

Discussion

Tay Sachs disease provides a model for mass screening of adults so that couples at risk to have a child affected with the disease can be identified, informed of their risks, and offered prenatal diagnosis. While virtually every medical student knows about the classic cherry red spot (6), the medical community as a whole has failed to inform patients of the facts and encourage participation in screening programs (7, 8, 9). Our study population performed reasonably well with regard to objective knowledge about Tay Sachs disease. Pediatricians and neurologists were the most knowledgeable but this is probably true because they most often see the affected children. The poorer results of internists and obstetrician-gynecologists were of greater concern since they are the primary physicians of the at risk parent population.

It is our feeling that the population surveyed probably scored higher than might be expected in the general medical community because of the long association of this institution with Dr. Sachs who first described and continued to study this disease (10, 11).

In our study, the most striking deficiency of knowledge was with regard to carrier incidence in the at risk population (6). Using Rosenstock's criteria, performance on this particular question proves especially significant. If one's perception of one's own susceptibility is minimized, motivation for compliance with screening is diminished. It follows that a physician who is unaware of the incidence of the disease cannot effectively serve as an advocate to encourage patients to participate in screening.

While the construction of the question assessing knowledge of the mode of inheritance in Tay Sachs disease did not allow us to evaluate the entire population, it is evident that a substantial number of the individuals surveyed do not have an adequate grasp of this material. Over 20% of those who knew how Tay Sachs was transmitted could not translate this information into the correct risk figures. This finding is disturbing and points to the necessity for increased emphasis on the basic modes of inherit-

TABLE III

Performance of Physician Population on Specific Questions Pertaining to Tay Sachs Disease

Aspects of Tay Sachs	Percent Physician Population Responding Correctly			
	MS	HS	ATT	Total
Clinical Manifestations	92.4	92.4	88.0	91.5
Ethnic Preponderance	83.3	85.6	88.0	85.5
Mode of Inheritance	90.9	82.2	84.0	85.0
Prognosis	81.8	81.4	96.0	84.6
Etiology	71.2	79.7	78.0	76.9
Incidence of Carrier State	39.4	31.4	40.0	35.5

MS = Medical Students; HS = House Staff; ATT = Attending Staff.

TABLE IV

Performance of 199 Respondents (85%) Who have Correctly Determined Mode of Transmission of Tay Sachs on Questions Concerning Risk of Carrier Parents Having Affected Child

Correct Response to		
Risk of Having Affected Child With Two Carrier Parents	Risk of Having Affected Child With One Carrier Parent	Both Responses
160	173	158
80.4%	86.9%	79.4%

TABLE V

Participation in Tay Sachs Screening Programs

Physician Subgroup	Number at Risk	Number Tested	Percent Compliance
Married	101	16	15.8
Single	71	8	11.3
House Staff	91	16	17.6
Attending Staff	42	4	9.5
Medical Students	39	4	10.3
Pediatrics	23	7	30.4
Neurology	14	1	7.1
Obstetrics/Gynecology	12	3	25.0
Medicine	21	3	14.3
Ophthalmology	19	1	5.3
Surgery	33	5	15.2
Psychiatry	11	0	0.0
Total	172	24	13.9

ance throughout medical school and post graduate education.

On the basis of our data, we believe that an educational program for medical staff is warranted with the intent of disseminating specific information about Tay Sachs disease and the availability

of screening. A major priority is widespread public education associated with screening programs must be to reach the medical community. If physicians are informed, they can actively support screening programs and hopefully help to lower the incidence of this tragic and fatal disease.

TAY-SACHS DISEASE QUESTIONNAIRE

Instructions: Please respond to the following questions by checking one or more appropriate answers.

1. Sex: male____ female____
2. Marital Status: married____ single____ engaged____ widowed____ divorced____ separated____
3. Parental Status: no children____ one or more children____ pregnancy underway____
4. Religious Origin: Jewish____ Roman Catholic____ Protestant____ Other____
5. Religious Origin of Spouse (if married): Jewish____ Catholic____ Protestant____ Other____
6. Hospital Staff Status: Attending____ Intern____ Resident____ Fellow____ Medical student____ Other____
7. Medical Specialty: Medicine____ Surgery____ Pediatrics____ Neurology____ Ophthalmology____ OB/GYN____
Psychiatry____ Other____
8. Tay-Sachs disease is a genetic disorder caused by a:
a) chromosome abnormal- b) biochemical defect____
ity____
9. The mode of inheritance in Tay-Sachs disease is:
a) autosomal dominant____ b) autosomal recessive____
c) X-linked recessive____ d) other____
10. The population at risk for Tay-Sachs disease is:
a) Askenazi Jews____ b) Sephardic Jews____
c) Blacks____ d) Italian Catholics____
11. The carrier rate in the at risk population is:
a) 1 in 10____ b) 1 in 30____ c) 1 in 200____ d) 1 in 600____
12. If both parents are carriers of the Tay-Sachs gene, the risk of having an affected child is:
a) 0%____ b) 25%____ c) 50%____ d) 100%____
13. If only one parent is a carrier of the gene, the risk of having an affected child is:
a) 0%____ b) 25%____ c) 50%____ d) 100%____
14. The major clinical manifestations of Tay-Sachs disease involves:
a) skeletal malformations____ b) hematological abnormalities____
c) CNS function____ d) cardio-respiratory function____
15. The prognosis for a patient with Tay-Sachs disease is:
a) normal life span____ b) death in the newborn period____
c) death in early childhood____ d) death in adolescence____
16. Have you had a Tay-Sachs screening test?
a) yes____ b) no____
17. If you answered yes to question #16, where was the test performed?

18. If you have been tested, are you a carrier?
a) yes____ b) no____

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Acute Ulceration of Third Portion of Duodenum with Fatal Hemorrhage

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Abstract

Two cases of acute huge ulceration of the third portion of duodenum with fatal gastrointestinal hemorrhage caused by carcinoma and suppurative pancreatitis in the uncinate process are reported. The important anatomic and pathologic relationship between the third portion of duodenum, portosplenomesenteric vascular system and the uncinate process is described.

Introduction

The third portion of duodenum lies below the head of the pancreas and its uncinate process and the superior mesenteric artery and vein cross these structures ventrally. A disease process such as infiltrating carcinoma or suppurative pancreatitis in the uncinate process may extend to the adjacent blood vessels and duodenum and cause a huge ulceration of the duodenum with massive hemorrhage. The authors are reporting two such cases and emphasize the clinicopathologic importance of this anatomic region.

Case Reports

Case I

A 52-year-old white female was admitted to the Methodist Hospital for hematemesis and back pain. She had had back pain for a year and colonic diverticulitis had been diagnosed in the past. Two and half months prior to the last admission, she entered another hospital for acute abdominal pain. Exploratory laparotomy was performed and infarction of the proximal 35.0 cm. of the jejunum, thrombosis of small mesenteric veins and mesenteric abscess were found. The infarcted bowel and abscess

were resected. After discharge, she improved but still had back pain for which she took aspirin or Darvon^(R). During the few days prior to the last admission, her back pain became worse. She took six aspirin tablets on the day of admission and started to vomit fresh blood. She was not an alcoholic and had no history of peptic ulcer.

On admission, she was pale, in profound shock, and was not icteric. There were mild epigastric tenderness and increased bowel sounds. The pertinent laboratory data were: WBC 17,000/cu mm; hemoglobin, 5.7 g/dl; hematocrit, 16.7%; SGOT, 952 mU/ml (normal 7-40); lactic dehydrogenase, 1,674 mU/ml (normal 100-225); and total bilirubin, 1.2 mg/dl; coagulation profile was normal. She was transfused with five units of packed red blood cells and two units of whole blood. Because of the suspicion of acute hemorrhagic gastritis due to aspirin, cold saline irrigation of the stomach through a nasogastric tube was done. Throughout the hospital course she was hypotensive with a blood pressure around 80/60 mmHg and continued to bleed from the nasogastric tube. She died 36 hours after admission.

At autopsy (A77-19), the stomach and entire intestine were filled with clotted and fluid blood. A huge punched-out ulcer measuring 4.0 x 4.0 cm was found in the third portion of duodenum with necrotic pancreatic tissue at the base of the ulcer. Many denuded branches of the superior mesenteric blood vessels were also seen at the ulcer base. A rupture of the inferior pancreaticoduodenal artery was demonstrated (Fig. 1). The lumen of the superior mesenteric vein was interrupted by scar with

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Fig. 1. Case I—A huge undermined ulcer (solid arrows) in the third portion of duodenum with a denuded artery (probe and hollow arrow) at the base of the ulcer. Two white strings passing through the papilla of Vater (P) at the left upper corner.

thrombus at its proximal and distal portion. There was no gross tumor mass at the head of the pancreas. The soft tissue around the main trunks of the superior mesenteric artery and vein was indurated. Microscopically, there was infiltrating adenocarcinoma in the uncinate process of the pancreas with focal infiltration in the ulcer margin of the duodenum and the soft tissue around the trunks of the superior mesenteric vessels. The wall of the mesenteric and portal veins was also infiltrated by carcinoma. The papilla of Vater and adjacent pancreatic tissue were free of carcinoma. The pancreatic duct was patent. In addition, there were tumor metastases in the regional lymph nodes and a few small metastatic nodules in the hepatic capsule, diaphragm and pleura. The ruptured pancreaticoduodenal artery showed necrotizing arteritis with many gram positive cocci and a mural thrombus. Old thromboses of small arteries were also present at the ulcer base.

Case II

A 41-year-old white male was admitted for the second time to the Mount Sinai Hospital Services, City Hospital Center at Elmhurst because of abdominal pain, fever and weakness. The patient was a chronic alcoholic and had been admitted for the first time a year previously with complaints of periumbilical pain, nausea, vomiting, anorexia, weight loss and weakness of three to four weeks' duration. The abdominal pain was unrelated to food intake. On examination, he was febrile (38°C) and had tenderness in the periumbilical region and the right upper quadrant. The liver and spleen were not palpable. Laboratory examination revealed severe anemia with hemoglobin, 5.8 g/dl and hematocrit, 18%; blood chemistries were within normal limits. He was transfused. He became afebrile and his abdominal pain receded without specific therapy. Abdominal x-rays showed extensive calcification in

the pancreas and an upper gastrointestinal series revealed normal contour of stomach and duodenal bulb, but calcific spots were noted in the region of the head of pancreas with some stretching of mucosal folds in the duodenal sweep. A liver scan showed no evidence of cirrhosis. He was discharged asymptomatic ten days after admission with a hematocrit of 31% and was followed in the Alcoholic Clinic.

One year later, he was readmitted because of fever and abdominal pain of three weeks' duration. On examination, he was febrile (39.5°C) and had scleral icterus. There was generalized abdominal tenderness but no hepatosplenomegaly. Pertinent laboratory data were hemoglobin, 10.2 g/dl; hematocrit, 30%; WBC, 24,000/cu mm; BUN, 60 mg./dl.; total bilirubin, 9.6 mg/dl; sodium, 122 mEq/liter and serum amylase, 90 Dye U (normal up to 200). A blood culture grew *proteus mirabilis* and microaerophilic streptococci. He was started on intravenous ampicillin, 2 g every six hours with intravenous fluid and was also given aspirin suppositories for fever. On the second hospital day, his hematocrit dropped to 22% and dark red stools were noted. A nasogastric tube drained bright red blood that could be cleared with lavage. Ascites appeared on the third hospital day and paracentesis revealed yellow turbid fluid with many leukocytes and amylase, 110 Dye units. Culture of ascitic fluid was negative. He continued to bleed massively from the nasogastric tube and rectum. Multiple blood transfusions and vitamin K were given. His jaundice became deeper with total bilirubin, 15.0 mg/dl. Esophagogastroduodenoscopy was performed and the entire visualized mucosa from the esophagus to the second portion of duodenum was covered with blood; no bleeding source could be seen. He died on the fifth hospital day.

The autopsy (A73-59) showed very similar changes as seen in Case I, except that the ulcer was secondary to pancreatic lithiasis with suppuration. The entire gastrointestinal tract contained red blood. A huge punched-out ulcer containing a blood clot measuring 6.0 × 4.0 cm was seen in the third portion of the duodenum (Fig. 2). The base of the ulcer was formed by the pancreas. Small arteries had ruptured there. There was also destruction of the junction of the portal and superior mesenteric veins. The uncinate process of the pancreas was destroyed; the papilla of Vater was unremarkable. The pancreas was edematous and firm, and there was a large stone in the dilated pancreatic duct at the head of the pancreas and gritty whitish calcific material in the parenchyma of the entire pancreas. Mucopurulent material was seen in the pancreatic



Fig. 2. Case 2—A huge ulcer (arrows) in the third portion of duodenum filled with blood clot. The papilla of Vater (P) and opened common bile duct (B) at the top.

duct and a few abscesses were found in the head and body of the pancreas. There were also infected thrombi in the portal veins, a few liver abscesses and an infected saccular aneurysm measuring 1.0 cm in diameter in the mesenteric arterial trunk adjacent to the ulcer. The liver was not cirrhotic. Two liters of yellow turbid ascites was present. No malignancy was seen in the pancreas.

Discussion

Ulceration of the third portion of duodenum is rare. Besides the ulceration caused by suppurative pancreatitis and carcinoma of the uncinate process described herein, postbulbar peptic ulceration (1) and tumor ulceration of intestinal origin in the third portion of the duodenum have been reported (2, 3). One of the authors (Lin) also has seen an autopsy case of a radiation-induced bleeding ulcer in that portion that had not been diagnosed by repeated

upper gastrointestinal series and gastroduodenoscopy.

Two cases reported here show a characteristic gross appearance of the ulcer and its anatomic relationship with the uncinate process of the pancreas and portosplenomesenteric vascular system. A huge ulceration without fibrotic reaction in the base of an ulcer may indicate a rapid ulcerative process which can be partly due to an action of pancreatic enzymes and intestinal bacterial flora. Diagnosis of hemorrhage from this portion is hard to make by gastrointestinal x-rays. A selective arteriography certainly will help to locate the source of bleeding. A fiberoptic endoscope also can reach this portion, but a direct visualization of the ulcer may be difficult because of massive hemorrhage. Therefore, if one can exclude bleeding from the esophagus to the second portion of duodenum, exploratory laparotomy may be mandatory, although the surgeon may expect to encounter a situation with overwhelming odds against a cure.

Jejunal infarction has been reported as a complication of pancreatitis (4). In Case 1, the jejunal infarction was in retrospect a complication and also an initial clinical manifestation of carcinoma in the uncinate process of the pancreas. For this reason, during an operation for mesenteric thrombosis and infarction of the proximal jejunum, one should look for an underlying pancreatitis or carcinoma.

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Open-Door-Policy Pediatric Clinic: A New Approach

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Abstract

In an attempt to care properly for the pediatric population of the community surrounding Hospital for Joint Diseases and Medical Center, we established an "Open-Door-Policy" Pediatric Clinic. Our purpose was to provide a private practice environment that offered personalized, expeditious, continuity of care. The team approach to patient care was the focal point of management. The physical locale was redesigned to accommodate the anticipated needs. The results have been outstandingly positive. In less than one year, this innovative program increased patient visits by 500%. In 1975, there were 3000 patient visits. In 1978, there were 12,000, and in 1979, the anticipated number of patient visits will be 15,000 plus.

Introduction

Hospital for Joint Diseases and Medical Center (HJDMC) is situated on Madison Avenue at 123rd Street in the heart of East Harlem, New York City. The patient population is drawn mainly from East Harlem and the South Bronx. The ethnic composition is approximately 60% Black, 40% Hispanic, with less than 1% White. The community is severely medically handicapped because of many complex interrelated problems. The socioeconomic level is poor. Statistics drawn from pediatric charts indicate that seventy-five to eighty percent of our patients are medicaid patients. Five percent are union and partial payers, and 15% are treated at no charge because they cannot afford any payment at all. Additionally, the area suffers from a high incidence of crime, drug and alcohol abuse, unemployment, poor housing and ignorance. It is distressing to report that for the 150,000 residents of East Harlem, there are only 50 part-time private medical practices to serve them. Thus, when in need of medical attention, other than acute hospitalization, the population must seek help from hospital emergency rooms, clinics and medicaid mills. It is not surprising then, that in a neighborhood so riddled with problems and lacking in personalized medical care, that the infant mortality for East Harlem is three times

higher than any other area in New York City (New York City Department of Health Report).

It became clear to us at Hospital for Joint Diseases that, to meet truly the needs of the community, our approach to planning and treatment had to change. We needed to give a private practice environment to patients that would foster continuity of care, personal relations, prophylactic pediatric medicine, continuing medical education, sociopsychological well being, in addition to general and acute care.

Discussion

Physical Description

To accomplish our goals, it was necessary to redesign the clinic to be more autonomous in the medical care offered, and to improve patient flow. As illustrated in Figure 1, the total floor space of the clinic is 2500 sq. ft. There are six examination/consultation rooms, each occupying about 100 sq. ft. Each room can adequately accommodate parents, child, resident and pediatrician.

The walk-in treatment room is 125 sq. ft., and is equipped with oxygen, emergency trays, drugs, intravenous fluids and equipment for minor surgical procedures.

Waiting areas are arranged near examination rooms, with piped-in music to help make the waiting time pass more rapidly and more pleasantly.

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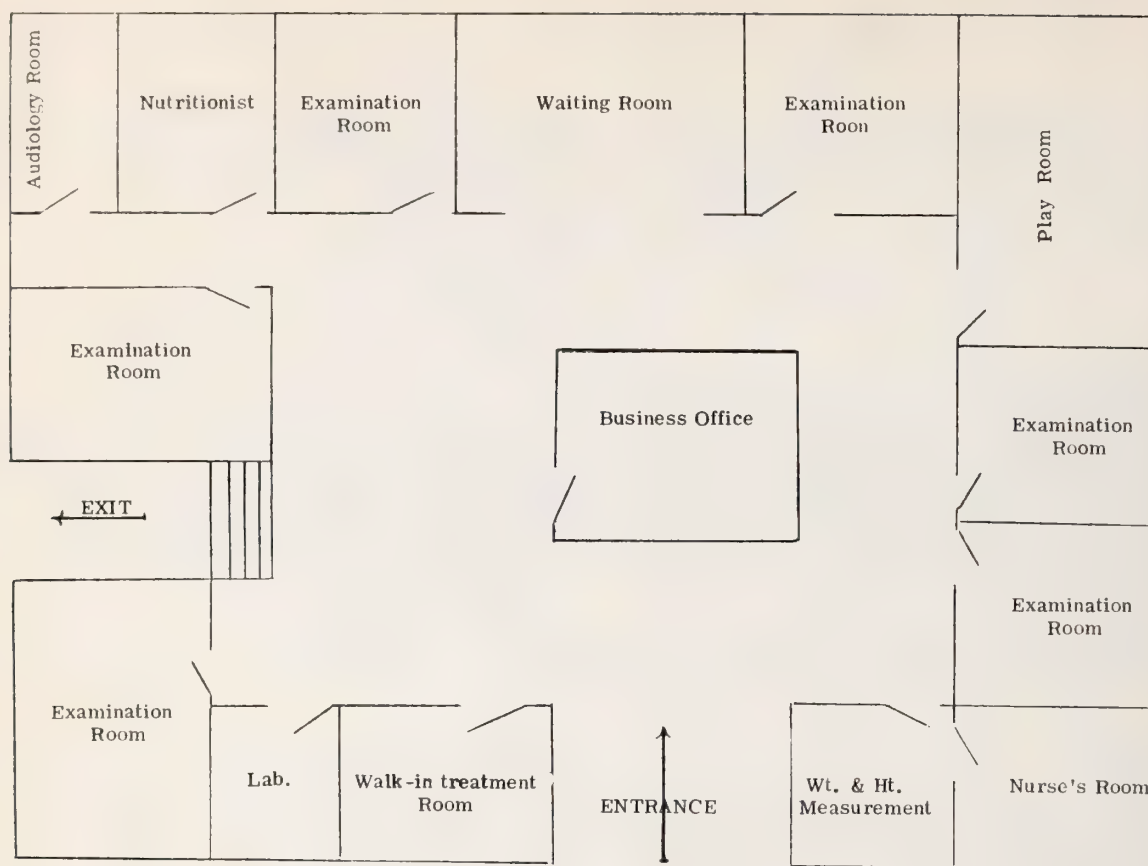


FIG. 1 Pediatric Clinic Hospital for Joint Diseases

Not far from the examination rooms is the 180 sq. ft. playroom, supervised by a fulltime playteacher and assisted by volunteers. (The playroom is used by a parttime pediatric psychologist for psychological evaluation of children during play.)

There is a nutritionist's room for private counseling of families which contains numerous pamphlets and leaflets to give out. Additional support is received through WIC (Women Infants and Children), a federally funded program for high-risk women with children who have nutritional problems. In off-peak hours, the nearby playroom is utilized for group sessions using slides and films as supportive educational tools.

The laboratory is stocked with micro- and macro-blooddrawing units, and equipment for urine analysis, hematology, and bacteriology. Other procedures are handled by the main hospital laboratory.

The clinic is further enhanced by a room equipped with an audiometer where noise levels are tested and, in the future, will contain a tympanometry apparatus. The same room is used for vision screening—far, near, and stereopsis. Finally, there

is a nurses' room used by a public health nurse, social worker, and staff nurses for private counseling of families, and a 120 sq. ft. business office with a registrar and all clinic charts.

Personnel Profile

Currently, there are six pediatricians appointed to the clinic. The clinic has one registered nurse, one licensed practical nurse, two nurses' aides, four nursing students, and one public health nurse who acts as a link between the hospital and the families through her visits for home evaluation. She also visits patients who fail to keep their appointments.

To round out the full medical care team, there are several others: 1) one nutritionist, 2) one laboratory technician, 3) one audiologist (a graduate student working under the supervision of the Director of the Audiology Laboratory at City College), 4) a parttime social worker, 5) a dental hygienist, 6) an eye screening technician (trained by the Ophthalmologic department at HJDMC), and 7) a sex educator for the adolescent clinic.

Management Plan

Clinic Hours: Clinic hours were expanded to offer service to families beyond school and work hours—8 a.m. to 8 p.m., Monday through Friday.

Patient Assignment: Families may choose their own pediatrician if they prefer. Names and hours of doctors are available in the business office of the clinic. Or, patients will be assigned to a pediatrician depending on how busy he is, with a maximum of 20 patients a day per pediatrician. This doctor, then, becomes the personal pediatrician for all the children in the family.

The clinic staff is divided into four panels. Each panel has one attending and one fellow, with one of them being available to the panel at all times. Although each physician has his own patients, the panel is familiar with all patients it covers. Thus, if a physician is absent or on rotation, the panel structure allows for patient coverage, continuity of care, and patient familiarity is not disrupted.

Registration and Appointments: Appointments are encouraged. Patients with appointments have priority on being examined first, unless there is a walk-in medical emergency. Every pediatrician has a direct telephone number in the hospital or patients may call the business office instead.

Every child below the age of 18 who comes to the hospital, will not go to the general registrar, but will be directed to the pediatric clinic where he is seen in the business office. Medical records are filed in the business office of the clinic, the chart being a subunit of the hospital chart with the same serial number. Charts are further identified by a color code according to panel. The patient is processed by the nurse, and goes through a medical examination at once. There is no waiting in line during registration. We have circumvented the length of time involved by having the patient's papers processed while the patient is being examined. In this way, we have managed to cut the length of clinic waiting time from an hour and half to one-half hour.

After Clinic Hours: Beyond clinic hours, a pediatrician is available in the emergency room to see patients 24 hours a day, seven days a week. It has been agreed that in every case and irrespective of seniority, the pediatrician in the emergency room should contact the patient's own doctor, discuss the case and arrange for the next visit if needed.

Additional Health and Referral Services

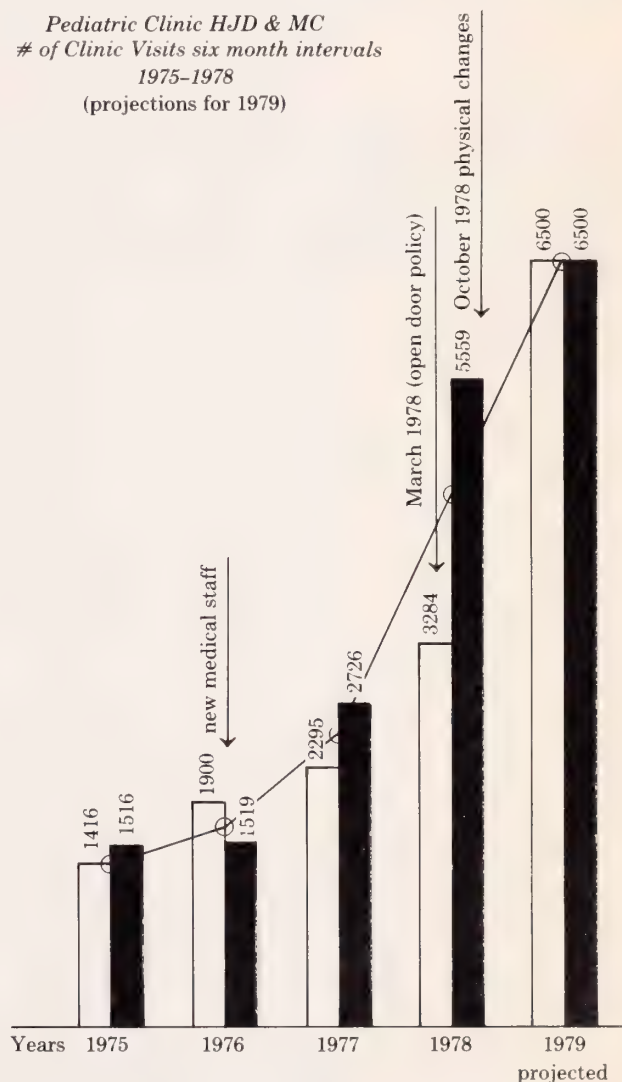
In addition to the medical acute care and health maintenance that the clinic strives for, arrangements have been made for urgent consultations.

The pediatrician can call the fellow or chief resident in pediatric orthopedics, pediatric oncology, surgery, and pediatric psychiatry. The consulting physician comes down to the clinic, examines the patient, and discusses the management with the patient's pediatrician.

For routine consultation, the patient is referred to the following clinics in HJDMC: 1) rheumatology, 2) dental, 3) ophthalmology, 4) dermatology, 5) otolaryngology, 6) radiology, 7) rehabilitation, 8) pain clinic, 9) pediatric orthopedics, 10) surgery, 11) alcohol and drug abuse clinic, 12) pediatric psychiatry, 13) neuromuscular clinic, and 14) nutritional clinic.

Arrangements have also been made for referral to the pediatric cardiology, diabetic, and hematology clinics at The Mount Sinai Medical Center. All cases are then referred back with consultation results. Patients may also be referred to the Audiology

Pediatric Clinic HJD & MC
of Clinic Visits six month intervals
1975-1978
(projections for 1979)



GRAPH I

Department, City College, for more comprehensive audiology studies.

Cases needing admission to the Hospital are admitted by their own pediatrician under the supervision of the attending in charge of the pediatric floor. Patients who need tertiary care, are referred to The Mount Sinai Hospital. Pediatricians are encouraged to see and discuss these cases with attendings at Mount Sinai.

Continuing Medical Education

Five hours a week are available for continuing medical education. Grand rounds are held once weekly in the pediatric department of HJDMC. Staff members are encouraged to participate as often as possible in the weekly grand rounds at Mount Sinai. Also a weekly requirement is the Journal Club at Hospital for Joint Diseases.

To further encourage efficiency and accuracy, the audit committee holds regular meetings and pediatricians are encouraged to attend. There are two official audits per year. Additionally, twenty clinic charts are reviewed by the pediatric group each month. Each pediatrician reviews his colleagues charts for deficiencies. This review process is conducted under the supervision of the director of the department.

Results

At first the staff had a difficult time adjusting to the new hours and management outlook since it varies considerably from most clinic routines. However, the highly enthusiastic response of patients and the team approach to patient management soon helped make the department a proud, consistently good team. Graph I illustrates the dramatic effect the "Open-Door-Policy" Pediatric Clinic has had on the community in number of patient visits. Based on statistics for the first four months of 1979, total patient visits at the end of the year will surpass 15,000.

At long last, a community hospital has achieved an efficient way of serving a poor local community. Eventually, statistics will be available as to the effect of continuity of health care, medical education, and personal attention that the patients have derived from this environment.

The system has been so successfully innovative, that it is being considered by several other community hospitals in similar neighborhoods. It is our hope that more and more institutions will adapt this policy in all clinics. In this fashion, institutionalized care will finally develop the outreach into the community that the community so desperately needs.

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Immunoblastic Lymphadenopathy with Hypercalcemia

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Abstract

Hypercalcemia, a recognized complication of lymphoproliferative disorders, has not yet been described with immunoblastic lymphadenopathy (IBL). We describe a case of IBL complicated by hypercalcemia in which a low immunoreactive parathyroid hormone level and a negative assay for osteoclast activating factor from the bone marrow and peripheral leukocytes are demonstrated. Bone marrow biopsy obtained prior to the onset of hypercalcemia revealed immunoblasts adjacent to areas of apparently active bone resorption.

IBL is a described lymphatic disorder (1, 2). We describe a case of IBL with two atypical features: the patient had a low IgA level and she developed hypercalcemia.

Case Report

A 59-year-old white female was first hospitalized in October, 1976 for anterior and posterior cervical lymphadenopathy. She was in good health until April, 1976 when she noted cervical lymphadenopathy. Physical examination revealed several, bilateral, 1 to 3 cm, nontender, firm nodes in the anterior and posterior cervical and submandibular regions. There was no hepatic or splenic enlargement. Peripheral blood cell counts and morphology were normal. Serum protein electrophoresis was unremarkable; no paraproteinemia or hyperglobulinemia was noted. Cryoglobulins were positive but were not characterized. A bone marrow biopsy showed two foci of mature lymphocyte aggregation with streaks of eosinophilic material on hematoxylin and eosin stain which was periodic acid-Schiff positive. These foci were not adjacent to bone spicules in the fields observed. A lymph node biopsy

revealed increased vascularity and eosinophilic deposition around blood vessel walls. The lymph node architecture was intact and no immunoblasts or eosinophilic deposition were found.

In January, 1977, she complained of a dry cough, pleuritic chest pain and an oral temperature elevation of 37.8°C. Blood and urine cultures were obtained and showed no growth. Physical examination revealed a 1 kilogram weight loss and persistence of the anterior and posterior cervical and submandibular lymphadenopathy as well as newly palpable, nontender, left axillary and left inguinal nodes. Liver span was 19 cm by percussion and the spleen tip was palpable 3 cm beneath the costal margin. Serum protein electrophoresis was again unremarkable. Immunoglobulin quantification showed an IgG 680 mg/dl (normal, 550 to 1900 mg/dl), IgA 27 mg/dl (normal, 60 to 330 mg/dl) and an IgM of 140 mg/dl (normal, 45 to 145 mg/dl). Coombs direct and indirect were negative. A bone marrow biopsy, a lymph node biopsy and a liver biopsy were obtained.

A serum calcium of 11.4 mg/dl (normal, 8.5 to 10.5 mg/dl) was observed on March 23 and three subsequent values were elevated. The serum albumin was 3.0 gm/dl (normal, 3.5 to 5.0 gm/dl), serum phosphorus was 3.7 mg/dl (normal 2.4 to 4.7 mg/dl), and alkaline phosphatase was 278 IU/ml (normal 30-90 IU/ml). She was treated with hydration, oral phosphates, and prednisone 30 mg on alternate days. The patient defervesced, remained afebrile and her calcium returned to normal. She was dis-

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charged March 27 on alternate day prednisone therapy of 30 mg.

She remained normocalcemic on prednisone therapy until June 26 when she was admitted with anorexia, spiking fever, dehydration and calcium elevations up to 13.8 mg/dl. The prednisone was discontinued on July 6. Serum calcium values as high as 10.2 with a simultaneous serum albumin of 2.1 gm/dl (normal, 3.5 to 5.0 gm/dl) were noted. On July 11, a bone marrow biopsy from the iliac crest was obtained and placed in tissue culture media for osteoclast activating factor assay determination. A plasma parathyroid hormone level was obtained. Prednisone was reinstituted and because of the deterioration of her clinical status, vincristine and cyclophosphamide were added. Serum calcium fell to 8.7 and fever abated with initiation of chemotherapy. Blood leukocytes were assayed for osteoclast activating factor two days later. The patient died on August 15.

Histopathology

Lymph Node of March 12, 1977 (Figure 1A and 1B): Microscopic studies revealed loss of normal architecture with rare isolated germinal follicles. Almost the entire node showed a sheet-like pattern of cells diffusely distributed without nodular aggregates. Prominent short branched vessels were seen throughout the lymph node, some with hyperplastic endothelial cells. High power reveals the cells to be lymphocytes, some plasma cells and immunoblasts with fairly clear nuclei, some of them with prominent small nucleoli and a moderate amount of cytoplasm. Occasional cells with abundant cytoplasm that appear vacuolated and eosinophilic are also seen. An occasional mitosis is seen. Foci of eosinophilic staining areas are seen between cells. Periodic acid-Schiff staining reveals a strongly positive substance with intracellular and perivascular deposition.

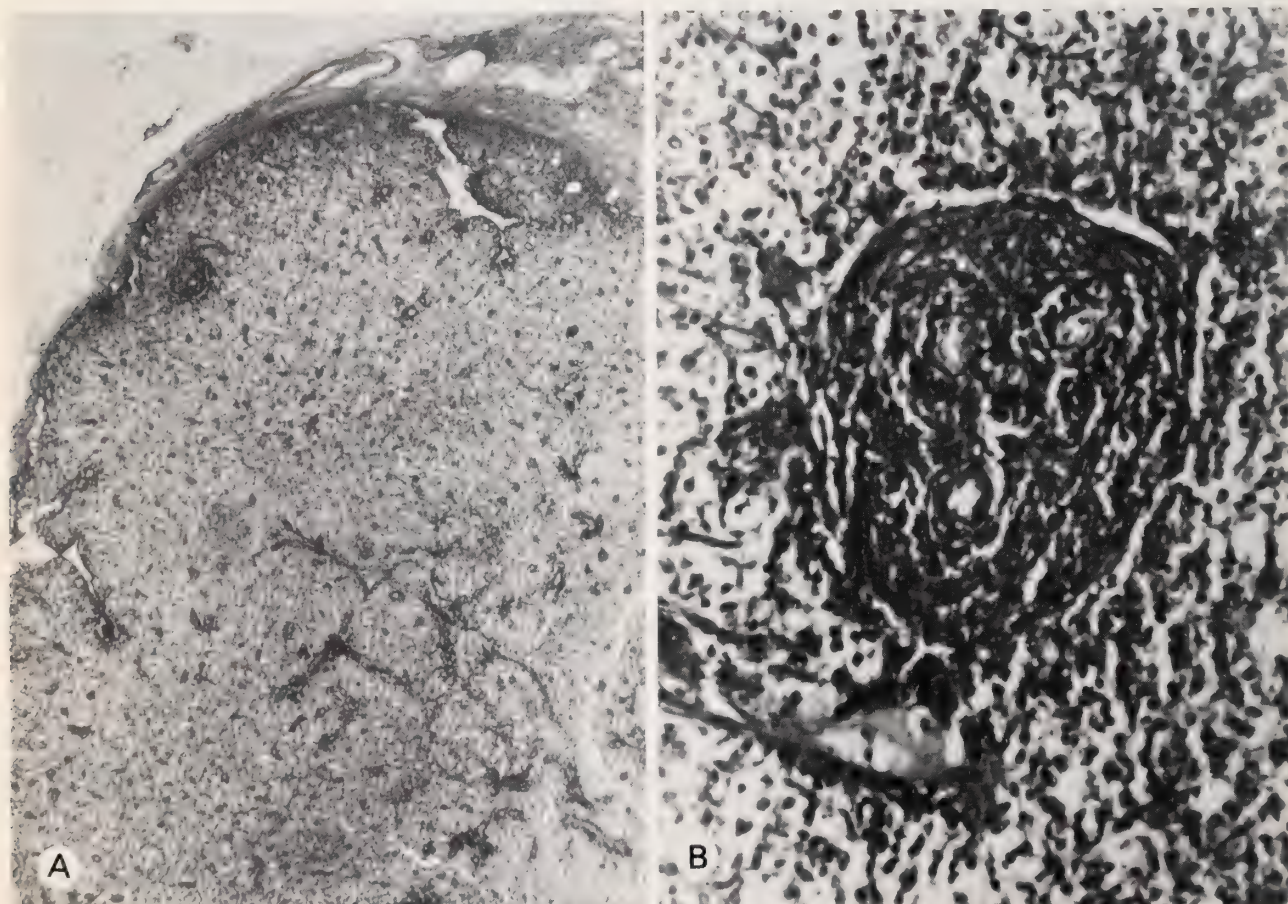


FIG. 1. Histology of involved lymph node, periodic acid-Schiff stain. A) Loss of normal architecture with prominent small vessel proliferation, 25 $\times$ magnification. B) High power view of hyperplastic blood vessel endothelial cells with intercellular deposition of periodic acid-Schiff positive material, 400 $\times$ magnification.

Bone Marrow Biopsies of March 21 and June 23, 1977: On microscopic examination under low power, a cellular marrow with a suggestion of a nodular pattern was seen. Hematoxylin and eosin revealed these nodules to have a marked eosinophilic staining background. The cell population consisted mainly of immunoblasts, lymphocytes and an occasional plasma cell. A periodic acid-Schiff stain was intensively positive and reticulum stain showed a marked increase in reticulum fibers lining the capillaries and small vessels within these nodules. In the bone marrow biopsy of March 21 obtained two days prior to the initial onset of hypercalcemia, the immune cells were noted to be adjacent to areas where obvious bone resorption was occurring. Intervening activated osteoclasts were not evident (Figure 2A and 2B).

Parathyroid Hormone

Parathyroid hormone levels were determined on blood samples obtained on July 8; serum calcium

was 9.9, phosphorus 2.6 and albumin 2.8. Using the Burrough Wellcome antibody (211/41) no detectable parathyroid hormone activity was measured.

Osteoclast Activating Factor

Osteoclast activating factor assay was performed by Dr. Gregory Mundy (4, 5) on July 9 and no significant activity was measured. Blood leukocytes were assayed for osteoclast activating factor (6) on July 14, two days after initiation of chemotherapy. No significant activity was observed.

Discussion

IBL was described as a lymphoma-like disease characterized clinically by fever, diaphoresis, weight loss, generalized or localized lymphadenopathy and hepatosplenomegaly. (1) (2) Associated findings may include hemolytic anemia, polyclonal hyperglobulinemia and cryoglobulinemia.

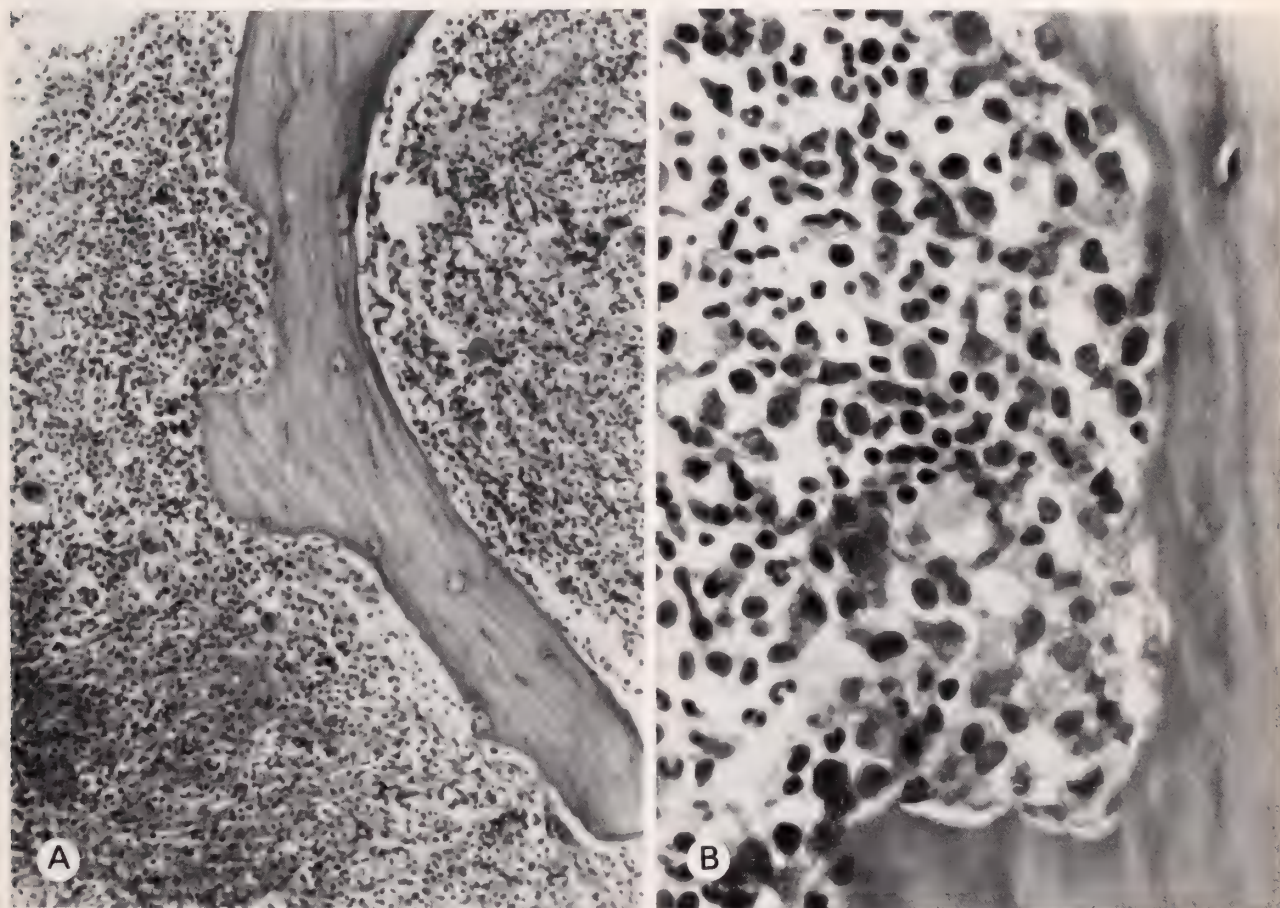


FIG. 2. Bone marrow biopsy taken two days prior to the initial onset of hypercalcemia, hematoxylin and eosin stain. A) Bone spicule demonstrating region of marked resorption with adjacent regions of immunoblast, lymphocyte and plasma cell invasion and eosinophilic material deposition, 100 $\times$ magnification. B) High power view of region of active bone resorption demonstrating contiguous immune cells including immunoblasts. Activated osteoclasts are not seen, 400 $\times$ magnification.

Our patient had recurrent fevers, diaphoresis, mild weight loss, generalized lymphadenopathy, and hepatic and splenic enlargement. There was no evidence of hemolytic anemia. A polyclonal hyperglobulinemia was not found; in fact, a low IgA was measured on immunoglobulin electrophoresis. This has been noted in three published cases (3). The patient also had a cryoglobulinemia that was not fractionated.

Hypercalcemia as a complication of IBL has not been previously reported. It is, however, a described complication of a number of malignancies including immune cell malignancies such as multiple myeloma, acute and chronic leukemia and Hodgkin's and non-Hodgkin's lymphoma (7). While the malignant cells may act directly on the bone matrix (8), evidence now points to the importance of humoral factors (9, 10) and host cells (11, 12) as agents of osteolysis.

We were unable to identify elevated parathyroid hormone or increased osteoclast activating factor as the cause of our patient's hypercalcemia. Coexistent primary hyperparathyroidism or ectopic hyperparathyroidism as a cause of the hypercalcemia was considered unlikely in view of the normal serum phosphorus, lack of roentgenographic as well as histologic bone changes and the unremarkable level of immunoreactive parathyroid hormone. Normalization of the hypercalcemia with prednisone and chemotherapy further argues against primary hyperparathyroidism. Osteoclast activating factor was not detected at the time of the assay, when measured in the peripheral blood or in the bone marrow. The bone marrow biopsies were examined for activated osteoclasts and none were observed. Instead, immune cells were seen to be adjacent to the sites of active bone resorption, suggesting cell-mediated bone lysis by the immune cells. Admittedly, review of bone marrow biopsy material obtained through the Vim-Silverman needle does not provide adequate material to estimate numbers of resorption centers or the predominate bone cells involved. Additionally, bone resorption has been shown to occur in the absence of activated osteoclasts (13). Nevertheless, the lack of activated osteoclasts and the pattern of immunoblasts adjacent to areas of active bone resorption was found consistently in the marrow samples taken prior to the onset of hypercalcemia. These differed from the

pattern found in earlier marrow specimens obtained when the patient was normocalcemic. The occurrence of hypercalcemia in IBL and the appearance of immunoblasts contiguous to regions of active bone resorption suggests that these cells may have in part caused the bone resorption and hypercalcemia that occurred.

Acknowledgement

The authors are indebted to Dr. Gregory Mundy for the measurement of osteoclast activating factor in the bone marrow and to Drs. B. Koziner and D. Strauss for their helpful comments.

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The Tarsal Tunnel

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Introduction

Although in recent years the tarsal tunnel syndrome has become a recognized clinical entity like the carpal tunnel, the anatomy of the tarsal tunnel has not been adequately elucidated to explain the symptoms. Since only nominal description of the structures below the medial malleolus and deep to the flexor retinaculum is found in standard textbooks of anatomy, the purpose of this paper is to document the pertinent surgicoanatomic relationships and the extent of the tunnel deep to the flexor retinaculum.

Materials and Methods

Forty feet were dissected. Prior to dissection, the following were noted: a) Center of the medial malleolus (M) b) Center of the heel (H). From the point (M) a perpendicular was drawn to the medial border of the foot (C). Another line was drawn from the point M to the midpoint of the heel (H). A third line was drawn from point M to point B which is the midpoint between H and C (Fig. 1). The line MB, used as the reference point for the bifurcation of the tibial nerve, closely approximated the distal border of the mid portion of the flexor retinaculum.

The skin of the distal anteromedial part of the leg and the foot was removed to expose the great saphenous vein, Tendocalcaneus, medial malleolus and part of the abductor hallucis muscle. By careful dissection, the entire flexor retinaculum was exposed.

The following measurements were taken after the removal of the retinaculum.

a) The point of bifurcation of the tibial nerve

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into the lateral and medial plantar nerves in reference to the midpoint of line MB.

b) The distance between the points of bifurcation of the nerve and the posterior tibial artery.

c) The distance of the first motor branch to the abductor hallucis from the point of bifurcation of the tibial nerve.

Cross sections of the leg and foot at 2.0 cm intervals were utilized for study, commencing about 6 cm above the medial malleolus to about 4 cm distal to it.

Observations

Careful dissection reveals that the flexor retinaculum is a fan-shaped band that consists of upper, middle and lower portions extending from the medial malleolus to the region of the heel and sole of the foot. The upper portion of the retinaculum is the thinnest and the direction of fibers is slightly obliquely upwards from the medial malleolus. This portion covers the tendons of the Tibialis Posterior, Flexor Digitorum Longus, Posterior Tibial vessels and nerve and is continuous superiorly with the transverse fascial septum that separates the deep muscles from the superficial muscles in the posterior compartment of the leg (Fig. 2). Inferiorly, this portion is continuous with the middle part of the retinaculum. The fibrous septae from the deep aspect of this portion of retinaculum are thin and not very conspicuous. The anatomic tarsal tunnel begins at the proximal border of the upper portion of the retinaculum and, at this point, the tendons and the neurovascular structures descent from the posterior compartment of the leg. Cross sections of the tunnel at this level indicate that the perineurovascular space is wide and is filled with fatty connective tissue (Figs. 3-5).

The middle portion of the retinaculum extends from the inferior part of the medial malleolus across the tendinous and the neurovascular structures to the distal part of the tendocalcaneus and the medial surface of the calcaneum. This portion is thicker



FIG. 1. Indicates the reference points on the medial side of the ankle.

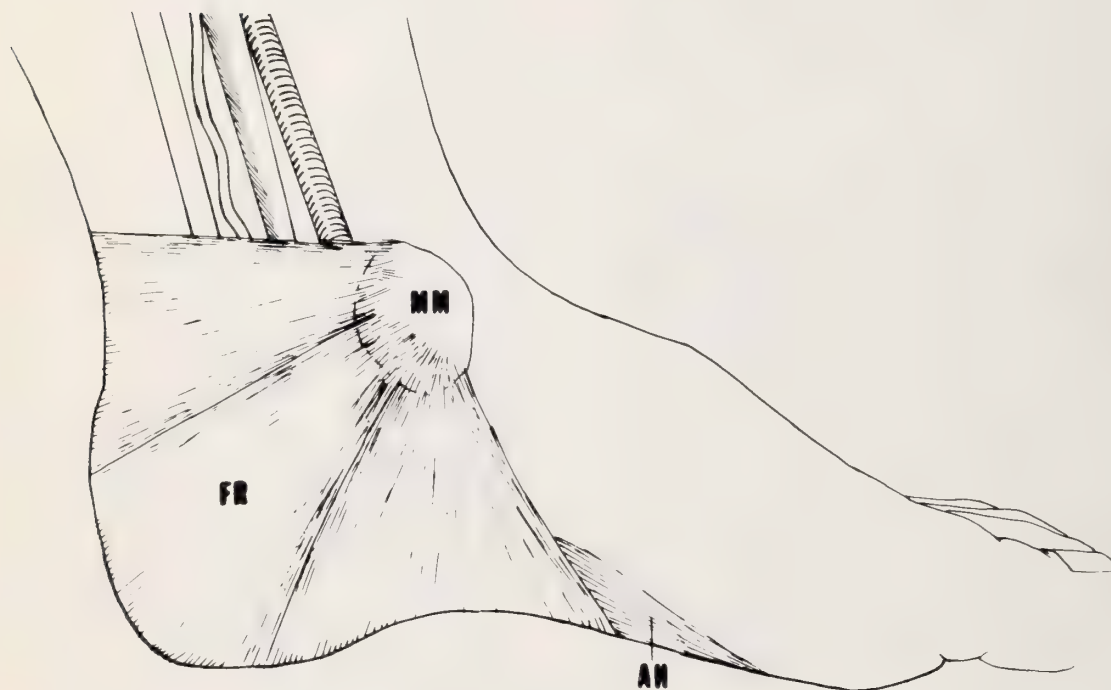


FIG. 2. The flexor retinaculum.

than the upper part and the septae from the deep aspect are continuous with those of the upper portion. Fibrous connections between the septae and the perineural connective tissue are prominent at this level. The tibial nerve bifurcates deep to this portion and the tarsal tunnel becomes narrow and splits distally into two tunnels for the lateral and medial plantar vessels and nerves (Fig. 6). The

upper distal part of this portion is perforated by the vein to the vena comites of the posterior tibial artery. The medial calcaneal vessels and nerve usually perforate this portion of the retinaculum.

The lower portion of the retinaculum is the thickest of the three and extends from the anteroinferior part of the medial malleolus, across tendons and the neurovascular structures to the medial surface

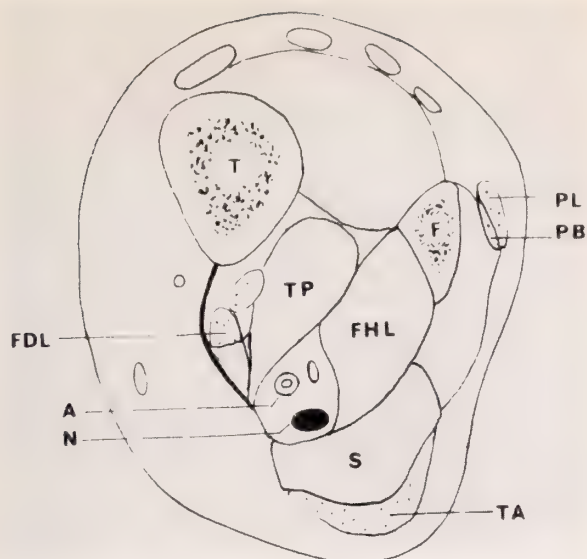


FIG. 3. Cross section of the leg denoting the proximal portion of the tarsal tunnel.

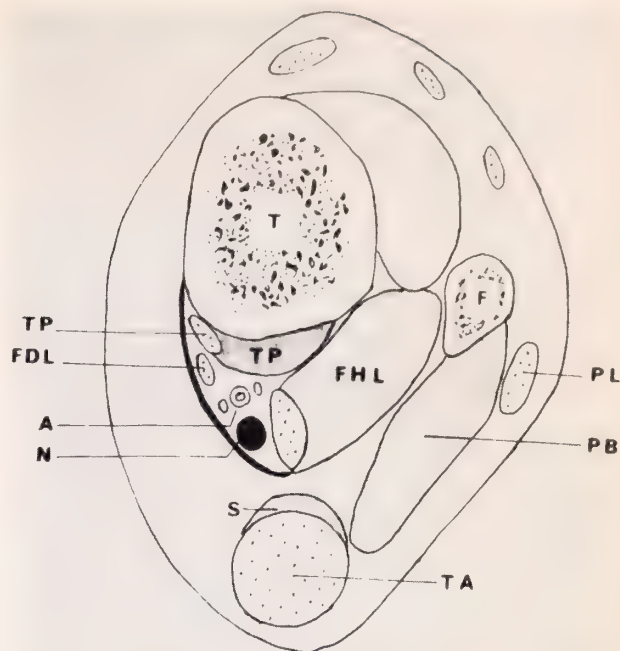


FIG. 5. Cross section of the leg about 2 cm above the ankle indicating the tarsal tunnel.

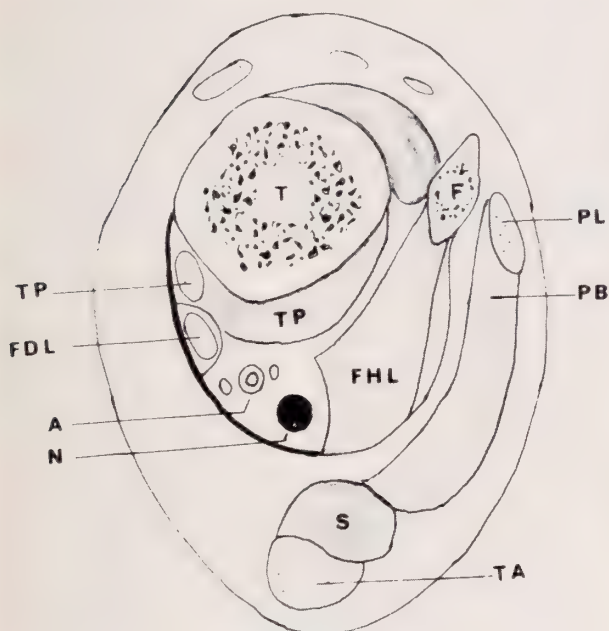


FIG. 4. Cross section of the leg showing the tarsal tunnel about 4 cm above the ankle.

of the calcaneus. This portion also blends with the plantar aponeurosis, fascia of the abductor hallucis and the deep fascia of the medial border of the foot distally. The fibrous septae of this portion are thick and blend with the deltoid ligament. The tarsal tunnel at this level is already split into two, to

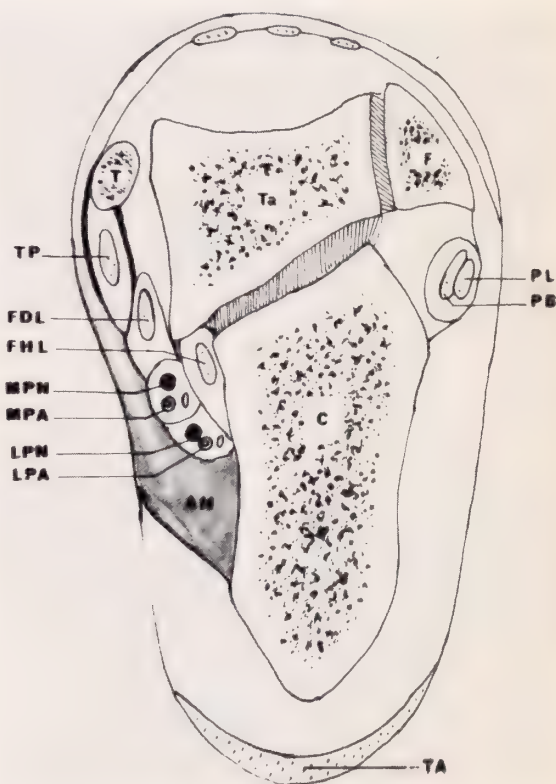


FIG. 6. Oblique section of the ankle showing the hallucis tunnel and the bifurcation of the tarsal tunnel.

accommodate the medial and lateral plantar vessels and nerves (Fig. 7, 8).

Removal of the flexor retinaculum revealed the following structures:

- 1) Tendon of the tibialis posterior

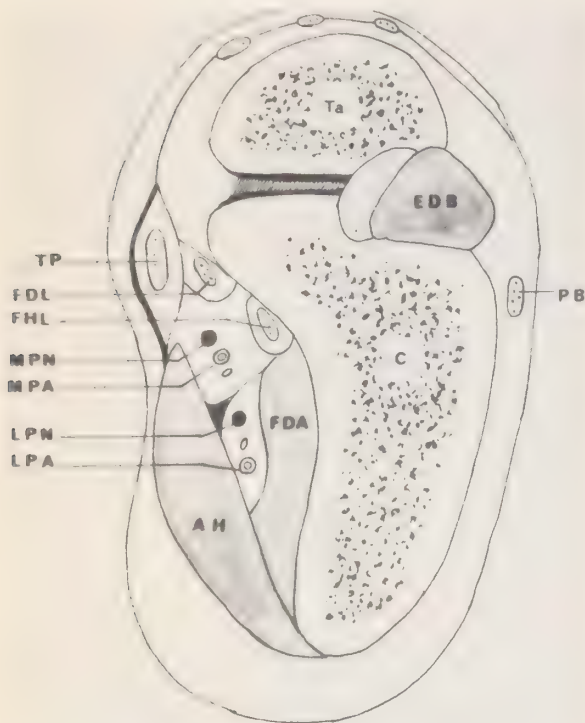


FIG. 7. Oblique section of the foot showing the boundaries of the upper and lower compartments of the tarsal tunnel.

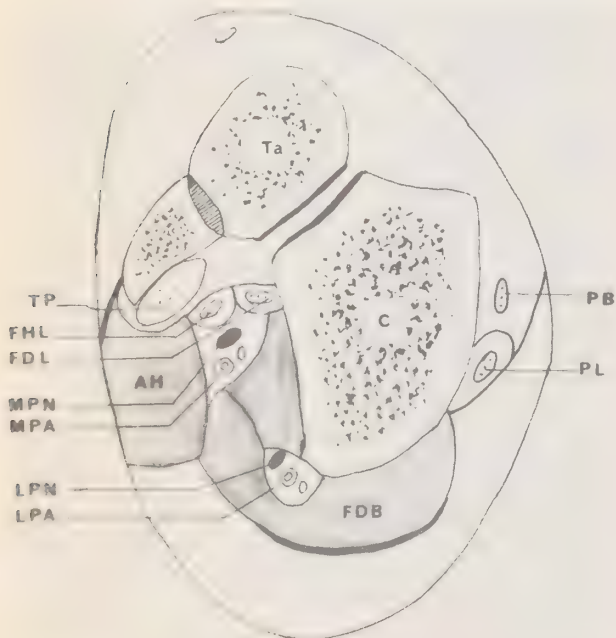


FIG. 8. Cross section of the foot showing the boundaries of the upper and lower parts of the tarsal tunnel distally.

- 2) Tendon of the flexor digitorum longus muscle
- 3) Posterior tibial vessels and nerve
- 4) Tendon of the flexor hallucis longus

These above structures occupy separate compartments deep to the flexor retinaculum (Fig. 9). Below the tendon of the tibialis posterior is the tendon of the flexor digitorum longus enclosed in its own synovial sheath. The tendon passes forward below the medial malleolus in a separate tunnel (Fig. 9), crosses the medial aspect of the sustentaculum tali and enters the sole of the foot by passing deep to the fibrous arch extending from the abductor hallucis to the navicular bone. The tendon is anterior to the neurovascular structures in the proximal part of the tarsal tunnel and superior in the distal part of the tunnel.

The posterior tibial artery is usually accompanied by a pair of vena comites that almost surround the artery completely. The vascular structures are enclosed in a fairly thick connective tissue sheath which is prominent up to the level of the inferior border of the malleolus. In about 25% of the specimens, the veins appeared dilated and varicose.

The posterior tibial artery descended posterior to the veins enclosed in a common fibrous sheath below the medial malleolus. At the point where the artery curves inferior to the malleolus, it gives off a small but constant articular branch to the ankle. Apart from its vascular function, this small branch also serves to anchor the main vessel at the medial

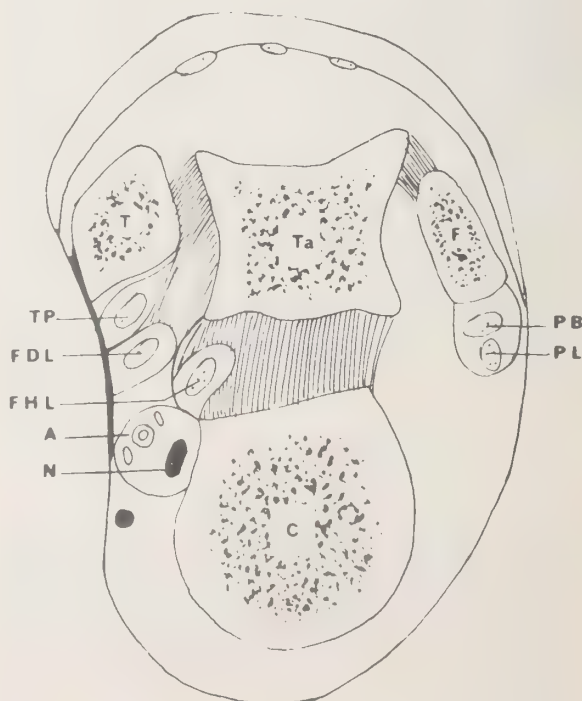


FIG. 9. Oblique section of the foot showing the distal part of the tarsal tunnel prior to bifurcation.

malleolus (Fig. 10). Beyond this anchoring point, the posterior tibial artery descended almost vertically over the lateral and medial plantar nerves forming an arterial arch and bifurcated about 1.8 cm distal to the bifurcation of the tibial nerve. It was noted that the bifurcation of the artery in 95% of the specimens was deep to the upper tendinous border of the abductor hallucis muscle and, at the point of bifurcation, the artery gives off a small tarsal branch (Fig. 10). Thus, the arterial arch seems to be fixed above by the anchoring artery and below by the tarsal branch, superficial to the nerves. The segment of the tibial artery above and below the anchoring artery was tortuous and slender branches from these loops could be traced to the perineural sheath. The lateral and medial plantar vessels follow their respective nerves to the sole of the foot (Fig. 10).

The tibial nerve, along with the blood vessels, is deep to the flexor retinaculum and insulated from the other structures by connective tissue derived from the retinaculum and by fatty connective tissue. The nerve descends along the medial side of the tendon of the flexor hallucis longus posterior to the blood vessels and the tendons of the flexor digitorum longus and the tibialis posterior in the upper part of the tarsal tunnel (Figs. 3-5). At the proximal part of the sustentaculum, the nerve is separated from the tendon of the flexor hallucis

longus by the medial wall of the hallucis tunnel which is formed by the downward prolongation of the fibers from the deltoid ligament superficial to the sustentaculum tali (Fig. 6). The nerve bifurcated deep to the middle portion of the retinaculum about 1.3 cm above the midpoint of the line MB into the lateral and medial plantar nerves. The medial calcaneal branch of the nerve was given off about 1.6 cm above the bifurcation of the tibial nerve. The lateral plantar nerve and vessels lie between the abductor hallucis medially and the quadratus plantae (FDA), laterally (Fig. 7). The nerve and the accompanying vessels pass to the lateral aspect of the sole of the foot by passing superficial to the flexor digitorum Brevis (Fig. 8). The medial plantar nerve and vessels lie in the upper division of the tarsal tunnel between the abductor hallucis and the medial wall of the hallucis tunnel which is inferior to the sustentaculum tali. Distally, the structures lie between the abductor hallucis medially and inferior to the tendons of the flexor hallucis and digitorum longus (Fig. 8).

The first motor branch from the medial plantar nerve to the abductor hallucis is given off about 3.4 cm from the bifurcation of the tibial. Distal to this motor branch, two or sometimes three branches were given off by the medial plantar nerve to the muscle.

The tendon of the flexor hallucis longus descends



FIG. 10. Denotes the anchoring artery (AA), tarsal branch (TB) and the bifurcation of the tibial nerve (N) above the abductor hallucis (AH).

along the posteromedial surface of the tibia and medial to the posterior tubercle of the talus. The tendon then passes inferior to the sustentaculum tali that forms the roof of a fibroosseous tunnel whose medial wall is formed by a downward prolongation of the fibers of the deltoid ligament (Figs. 6, 9).

Discussion

Though the tarsal tunnel syndrome has been established as a clinical entity, accurate definition and anatomical description of the tunnel is lacking. Many investigators have alluded that the flexor retinaculum (lancinate ligament) is a thin sheet of connective tissue extending from the medial malleolus to the medial surface of the calcaneum (1) and that the tarsal tunnel is the space occupied by the neurovascular structures deep to this thin sheet of connective tissue. However, it appears that this description refers only to the middle portion of the retinaculum and does not take into consideration the entire morphology of the retinaculum and the tunnel. This study suggests that the retinaculum is not a thin sheet of connective tissue but a fan-shaped band of varying consistency and thickness sweeping like an arc from about 10 cm above the heel to the distal part of the sustentaculum tali (Fig. 2).

The anatomic tarsal tunnel begins at the proximal border of the upper portion of the flexor retinaculum and extends posteroinferior to the medial malleolus to slightly beyond the sustentaculum tali. The tunnel consists of a proximal portion behind the medial malleolus, a middle portion below the malleolus and a distal portion anterior to the malleolus. Most of the medial wall (roof) of the tunnel is formed by the three portions of the retinaculum except in the distal part where the neurovascular structures are separated from the retinaculum by the intervening abductor hallucis muscle (Fig. 6). Contrary to the common description that the lateral wall (floor) of the tunnel is entirely osseous, in this study we found that only the middle portion of the tunnel has a bony lateral wall; whereas, in the proximal portion of the tunnel, the lateral wall is formed mainly by the long flexor tendons and the calcaneum.

The tarsal tunnel narrows distally, reducing the space-content ratio and divides into narrow upper and lower tunnels (Figs. 7, 8) for the medial and lateral plantar vessels and nerves, respectively. As depicted in Figure 6, the lateral wall of both tunnels is formed by the medial wall of the tunnel that

lodges the tendon of the flexor hallucis muscle. However, slightly distal lateral wall of the lower tunnel is formed by the flexor digitorum accessorius muscle and the upper tunnel is located between the abductor hallucis and the tendons of the flexor muscles (Fig. 7). In this position, both the medial and lateral plantar nerves could be subjected to ischemic neuropathy by the hypertrophy of the muscles. The proximity of the medial plantar nerve to the flexor tendons at this situation allows for the nerve to be implicated in inflammations of the synovial sheaths of the tendons or tumors of the tendons.

It has been documented that the tibial nerve bifurcates deep to the abductor hallucis muscle (1). However, in the present study, the level of bifurcation of the nerve was always above the superior border of the muscle and this fact facilitates the identification of the nerve for decompression which, in some cases, alleviates the pain that is the predominant feature of the syndrome. Though ischemic neuropathy has been blamed as the underlying cause of the pain and other early sensory symptoms (3) in more than 50% of the cases the precipitating factor for the ischemia is not obvious clinically and these cases tend to be classified as idiopathic.

In view of the unknown etiology, it is presumed that a minor lesion could alter the normal anatomic relationships of the nerve or its branches to its environment and precipitate the symptoms of the syndrome. As pointed out above, the anatomic factors that could trigger the onset could be muscular, fibrous, osseous and most likely, vascular. Compression of the nerve or its branches by dilated and engorged veins could be a causative factor in promoting the symptoms (4). Our studies indicate that the nerve and the vascular structures are much closer in the distal part of the tunnel than the proximal part and the presence of the vascular arch over the lateral and medial plantar nerves could be an insidious contributory factory to the causation of the syndrome. It should be emphasized that vascular factors are implicated in lesions like the carpal tunnel syndrome, trigeminal neuralgia and bells palsy of unknown etiology.

Recently, the tarsal tunnel syndrome has been associated with nontraumatic lesions such as rheumatoid arthritis (5), leprosy (6), ankylosing spondylitis (7) and space occupying lesions like neuroomas (8) and ganglia. The syndrome could be associated with fractures of the os calcis. These lesions constitute only a small percentage of the known etiologic factors of the syndrome.

Often difficult to identify and, therefore, overlooked is the medial calcaneal branch which is given off by the tibial about 1.6 cm above the bifurcation of the nerve. This nerve usually penetrates the midportion of the retinaculum and divides into branches to innervate the heel. It has been stated that this nerve is a branch of the lateral plantar nerve (9); though in this study, the nerve was constantly given off by the tibial. Similar to the lateral femoral cutaneous nerve which could be compressed by the inguinal ligament and produce meralgia paraesthetica, the compression of the medial calcaneal branch by the retinaculum would result in pain in the heel. This nerve should be identified and isolated during surgery because inadvertent denervation of the nerve would produce loss of sensation in the heel and medial aspect of the ankle which is an unwanted complication.

Acknowledgment

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Hypertrophic Osteoarthropathy Associated with A Malignant Thymoma

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Abstract

A patient is described who had symptoms of hypertrophic osteoarthropathy and was found to have a malignant thymoma. This is the second report of such an association and the first one suggesting that a malignant thymoma can cause hypertrophic osteoarthropathy.

Although thymomas have been associated with a vast array of clinical disorders and hypertrophic osteoarthropathy has been identified in patients with many different types of intrathoracic neoplasms, a search of the English literature reveals that the coexistence of the two in the same patient has only been reported once before (1). That report, which appeared in 1939, described a 14-year-old boy who died of a mediastinal tumor resembling a thymoma and during his illness had marked pulmonary osteoarthropathy. The present report describes a second patient with a malignant thymoma and hypertrophic osteoarthropathy and, for the first time, offers support for an etiologic relationship in that the boney changes disappeared after the neoplasm was removed surgically and the mediastinum was irradiated.

Case Report

A 52-year-old construction worker had a chief complaint of pain and swelling in the ankles for one month. He also complained of pain in the knees and wrists, swelling of the fingertips, occasional right precordial chest pain, and a ten pound weight loss. He gave a history of 30-pack-years of cigarette use and no occupational exposure to toxic or irritating substances. Physical examination was normal ex-

cept for marked clubbing of the fingers and toes and nonpitting ankle edema.

A posteroanterior chest roentgenogram revealed elevation of the right diaphragm and a right perihilar density (Fig. 1). A lateral chest roentgenogram demonstrated an anterior mediastinal mass or intrapulmonary density in the area of the right middle lobe (Fig. 2). Roentgenograms of the bones revealed new periosteal bone formation involving the distal radius, ulna, tibia, and mid-first metacarpal bilaterally (Fig. 3). Serum electrolytes and complete blood count were normal.

Fiberoptic bronchoscopy revealed extrinsic compression of the medial segment bronchus of the right middle lobe. Rigid tube bronchoscopy and mediastinoscopy were normal. At thoracotomy, a seven by eight centimeter irregularly lobulated mass was found in the anterior mediastinum that extended from the lower pole of the right lobe of the thymus (Fig. 4). It was adherent to the medial aspect of the right middle lobe of the lung and the adjacent pericardium and removal necessitated excision of the thymus, portions of the pleura in the area of the right middle lobe and portions of the pericardium. Light microscopy sections revealed nests of uniform epithelial-like tumor cells, divided by fibrous bands, that contained eccentric nuclei, vacuolated cytoplasm, and dark staining rims (Fig. 5). Sections taken at the periphery of the tumor showed that the cells invaded surrounding connective tissue and were intermixed with a few lymphocytes. Electron microscopy sections revealed that the cells were joined by desmosomes and contained tonofilaments, thus, confirming that they were epithelial in origin rather than lymphocytic (Fig. 6).

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FIG. 1. Admission chest roentgenogram showing elevation of the right diaphragm and right perihilar density.

Following recovery from surgery, the patient received 4,500 rads of irradiation to the mediastinum over a three-week period. During the subsequent months, the pain in the wrists, ankles and knees disappeared, the clubbing resolved, and bone roentgenograms returned to normal (Fig. 7).

Discussion

Our report documents the association of hypertrophic osteoarthropathy with a thymoma and suggests a causal relationship. A previous report of such an association described a 14-year-old boy who

died of an invasive mediastinal tumor that could not be classified microscopically but resembled a carcinoma rather than a sarcoma (1). The microscopic descriptions provided in the text suggest that, by using current classifying criteria, the tumor was probably an epithelial-cell thymoma. In our patient, gross pathology and light and electron microscopy findings were diagnostic of a malignant thymoma of an epithelial-cell type.

The classification of thymomas has been an area of controversy for years, particularly since early authors classified most thymic tumors, including lymphomas and lymphosarcomas, as thymomas



FIG. 2. Lateral chest roentgenogram showing anterior fullness in the area of the right middle lobe.

largely on the basis of anatomic position. More recently, primary tumors of the thymus have been classified histologically on the basis of the predominant endogenous cell present (epithelial cell or lymphocyte) and other tumors involving the thymus of nonthymic cell origin have been excluded from the classification (2). Even then, however, true thymomas may be difficult to recognize and classify because they can assume a variety of microscopic patterns, including most commonly spindling, pseudo-rosette formation, or vacuolization, and can

contain variable combinations of epithelial cells and lymphocytes. Because of these nosological difficulties, electron microscopy has become an important tool in not only differentiating thymomas from other tumors invading the thymus but also in allowing recognition and separation of epithelial cells and lymphocytes on the basis of the presence or absence of desmosomes and tonofilaments (3).

Although patients with epithelial thymomas tend to have a poorer prognosis than those with tumors predominantly containing lymphocytes, the most

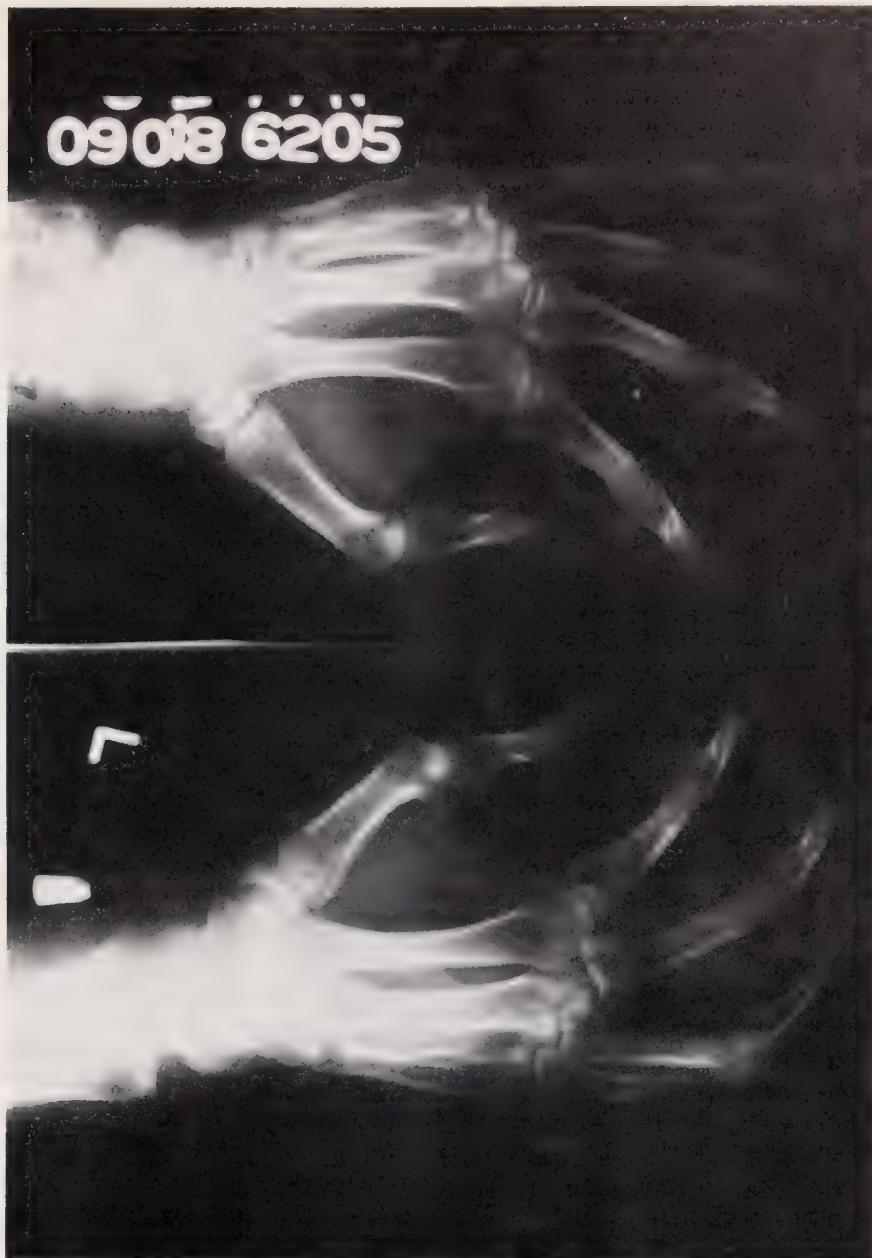


FIG. 3. Roentgenograms of the hands showing cortical changes along the inner aspect of the first metacarpal bones.

important factor affecting survival is not the cell type but the extent of local invasion (4). Those tumors confined to the thymus are considered benign and are associated with a good prognosis; whereas, those locally or distantly invasive are considered malignant and are associated with a poorer prognosis. Since invasive ones tend to grow slowly, with rare metastasis, and can potentially cause death by extensive local infiltration of the mediastinum, surgical resection is the primary form of therapy. Radiotherapy is also effective therapy and should be administered to patients with unresecta-

ble tumors and probably also to those with invasive tumors successfully removed surgically, although the indications for radiotherapy in the latter group are less clear (5).

Hypertrophic osteoarthropathy has been associated with many different types of benign and malignant intrathoracic tumors (6). Peripheral tumors involving pleura, particularly mesotheliomas, are more frequently associated with hypertrophic osteoarthropathy than central lesions, suggesting that involvement of pleura is an important etiologic factor (7). This is supported by findings that some



FIG. 4. Photograph of the gross specimen showing the thymoma extending from the right lobe of the thymus.

nonmalignant intrathoracic processes, such as esophageal dilation, neurofibromas, or dermoid cysts of the mediastinum that do not invade lung but do extend to pleural surfaces are also occasionally associated with hypertrophic osteoarthropathy (7). These findings, however, are difficult to correlate with observations discussed in this report that thymomas very rarely cause hypertrophic osteoarthropathy and yet frequently invade pleura and mediastinal structures.

In the patient described herein, the disappearance of subjective and objective findings of hypertrophic osteoarthropathy following surgery and irradiation strongly suggests that the original complaints and bony changes were related to the neoplasm. Therefore, considering that the treatment and prognosis are different for patients with thymomas as compared to those with bronchogenic carcinomas, thymomas should be considered in the differential diagnosis of patients with anterior me-

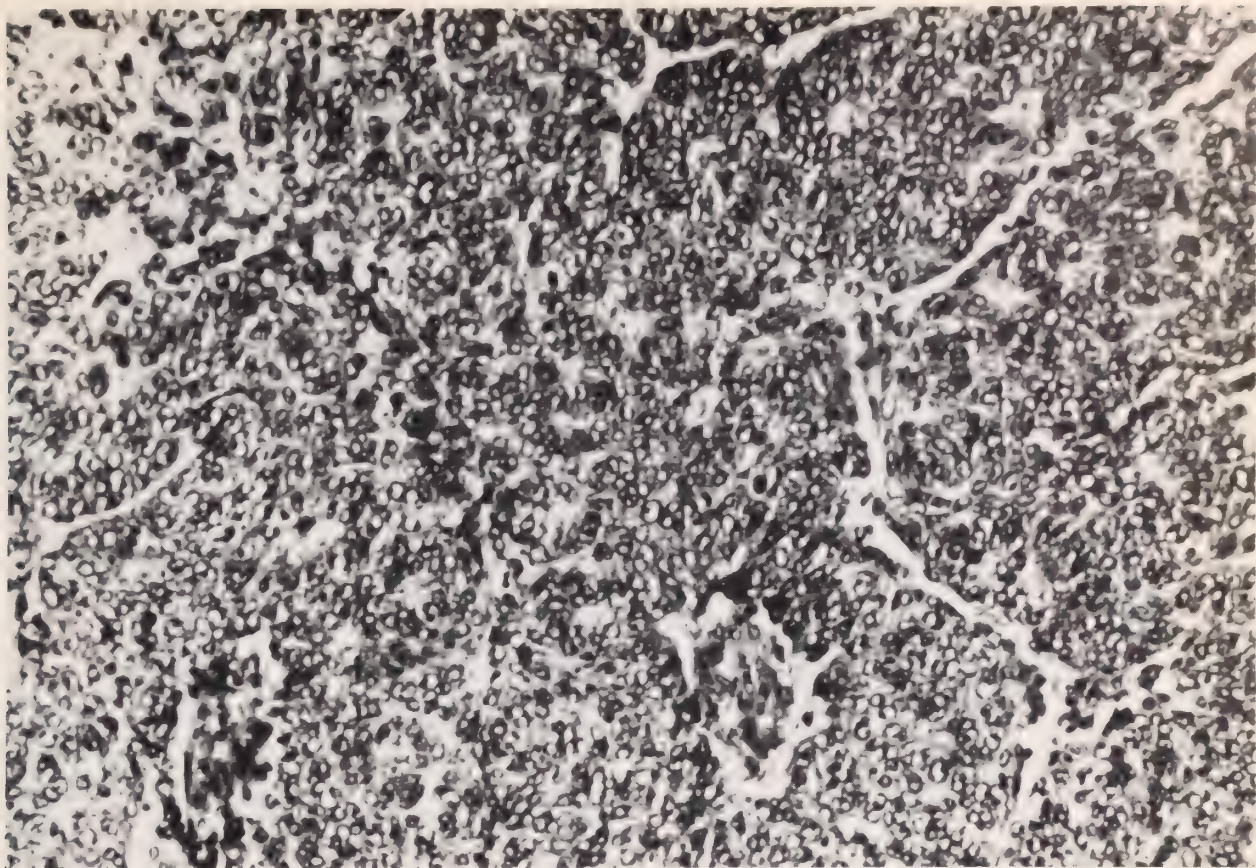


FIG. 5. Photomicrograph of the tumor showing uniform vacuolated cells (hematoxylin and eosin stain; original magnification $\times 200$).

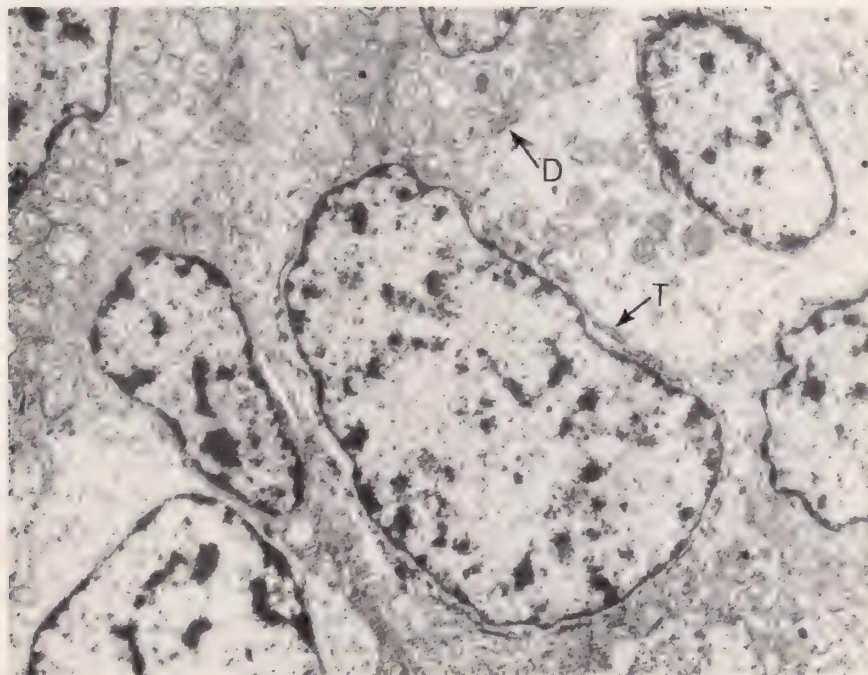


FIG. 6. Electron photomicrograph showing several cells and desmosomes (D) and tonofilaments (T) (origin magnification $\times 20,000$).

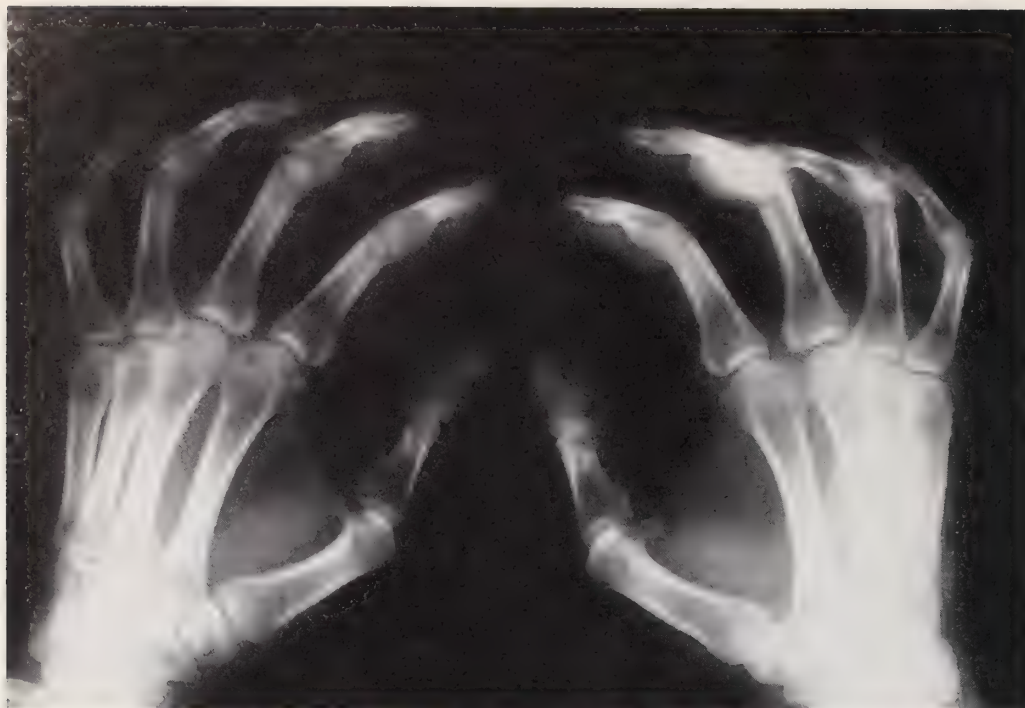


FIG. 7. Roentgenograms of the hands taken two months after removal of the tumor showing disappearance of cortical changes along the first metacarpal bones.

diastinal masses or apparent anterior intrapulmonary densities seen on roentgenograms who also have hypertrophic osteoarthropathy.

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Cecal Infarction: Ergot Abuse as a Possible Etiologic Factor

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Abstract

A case of localized cecal infarction in a thirty-year-old man with signs and symptoms of acute appendicitis and a long history of drug abuse is reported. Ingestion of unknown, but large amounts of ergotamine and intravenous injection of dissolved hydromorphone tablets prior to admission are discussed as possible etiologies of this patient's unusual pathologic findings. Gastrointestinal effects of ergot-containing compounds are discussed.

Introduction

Intestinal infarction is usually not present in young individuals unless associated with a congenital anomaly, an incarcerated hernia, or an obstruction caused by previous surgery. Pseudoobstruction of the intestine has been associated with drug abuse (1). However, to our knowledge no reports of intestinal infarction related to drug abuse exist (2, 3). Ergotism, secondary to ingestion of contaminated rye was seen in widespread epidemics during the dark and middle ages. Today, ergotism is almost always associated with the treatment of migraine headache. Very few reports of adverse effects of ergot compounds on the bowel exist (4-6).

We present an unusual case of localized cecal infarction unrelated to appendicitis, and a possible relationship to ergot ingestion and/or drug abuse.

Case Report

A 30-year-old white man was admitted to the Veterans Administration Medical Center at Newington with an 18-hour history of mild epigastric abdominal pain that had localized in the right lower quadrant and increased in severity during the four to five hours prior to admission. The patient re-

ported nausea, but denied vomiting and diarrhea. Several years prior to this admission, he had undergone an upper gastrointestinal series and endoscopy that were reported as normal in the workup of vague epigastric pain. There was no history of any urinary tract symptoms. A complicated social history was elicited involving heroine addiction for which he had undergone partial detoxification approximately six years prior to admission.

He freely admitted to taking large quantities of propoxyphene (Darvon) and other analgesic preparations. The patient also gave a history of migraine headaches and reported episodic use of ergot-containing compounds in large quantities. He reported taking large amounts of ergotamine 72 hours before admission, but on subsequent postoperative questioning, reported taking the drug 96 to 120 hours prior to admission. He also admitted that he had dissolved several hydromorphone (Dilaudid) tablets in water and administered this preparation intravenously.

On admission, the patient had a temperature of 102.8°, blood pressure 110/60, and pulse 105. The patient was an ill-looking young man in obvious distress, who appeared highly nervous and agitated. Examination of head, eyes, ears, nose, throat and chest were normal. Examination of the abdomen revealed generalized tenderness in all quadrants with severe right lower quadrant tenderness, guarding and rebound. Rectal examination was normal. There were no audible bowel sounds. The rest of the physical examination was within normal limits.

Laboratory work on admission revealed a hemoglobin of 12.5 gm., hematocrit of 37.5%, WBC 12,500

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with a differential shift to the left. Urinalysis revealed 10 to 15 white cells per high power field and had a specific gravity of 1,035. The impression on admission was ruptured appendicitis.

The patient was prepared for surgery with appropriate fluid and antibiotic therapy and underwent abdominal exploration through a standard McBurney incision. Upon entering the abdominal cavity, it was apparent that there was a marked amount of thick, grayish, exudative material in the right lower quadrant. Bowel contents were not observed. The appendix was normal. The cecum was delivered through the operative wound and there was an edematous, gangrenous patch, 4 cm in diameter, on the antimesenteric cecal wall, several centimeters from the base of the appendix (Fig. 1). There was no obvious perforation nor was a diverticulum present. The incision was extended medially across the midline and further abdominal exploration was performed with no other pathologic findings. The internal inguinal ring was found to be of normal size by palpation and inspection. Transversalis fascia was intact and there was no evidence of a hernia defect in the anterior or posterior pelvis. The terminal branches of the superior mesenteric artery including the ileocolic and all its branches were found to be freely pulsatile with no evidence of any vascular pathology. The area of gangrenous cecum was resected, and an ileocolic resection was performed. There was good bleeding from the transected bowel margins. The proximal end of the cecum was oversewn, and an end-to-side anastomosis of end intestine to the side of the ascending colon was performed. Copious irrigation of the right lower quadrant was performed. The abdomen was closed in standard fashion after two drains had been

brought out through a separate stab incision. Aerobic and anaerobic cultures of the peritoneal cavity were reported as no growth. Smear showed many polymorphic nucleocytes. Postoperative course was uneventful and unremarkable.

On pathologic examination, the distal ileum and appendix were grossly normal. The cecum measured 10 cm in length and 6 cm in diameter and was reddish in color except for a protruding dome-like area of the anterior wall 4 cm in diameter which was brown-gray in color. The wall was thin but intact and the mucosa was granular and yellow contrasting to the reddish color of the remaining cecal mucosa.

Microscopic examination of the cecum revealed necrotic mucosa with bacterial colonies. The submucosa was densely infiltrated by polymorphic leukocytes and showed dilated blood vessels, occasional fibrin thrombi and focal hemorrhages. At the center of the lesion, the muscular layers were destroyed and the purulent infiltrate extended to the serosa. Microscopic examination of the appendix was unremarkable. Final microscopic diagnosis was localized infarct of the cecum with secondary acute polymorphonuclear inflammatory infiltration.

Discussion

Ergot alkaloids stimulate smooth muscle contraction, central sympathetic activity, and peripheral adrenergic blockade (7). The smooth muscle contraction is most readily observed as uterine contraction and peripheral vasoconstriction, the latter of which is caused by stimulation of alpha receptors to which these drugs are tightly bound. Intense vasoconstriction resulting in ischemic gangrene of the extremities was one of the major hallmarks of the great European pandemics caused by ergot-contaminated rye (7). Numerous reports of reversible and irreversible peripheral ischemia with resulting gangrene are reported in the medical and surgical literature. Some reports document the peripheral vasoconstriction with arteriographic as well as noninvasive techniques (5-8). Renal ischemia has also been reported (9).

Central nervous system effects of ergot are complicated and varied. They may be manifested by a variety of symptoms including convulsions, clonus, pain, weakness, paraplegia, ophthalmologic conditions, etc., all of which or may not be permanent. In addition, a wide variety of mental disturbances and aberrations have been reported (7). Intestinal manifestations of ergot intoxication are rare. Stillman reported a case of ischemic colitis related to oral ingestion of ergotamine documented by barium

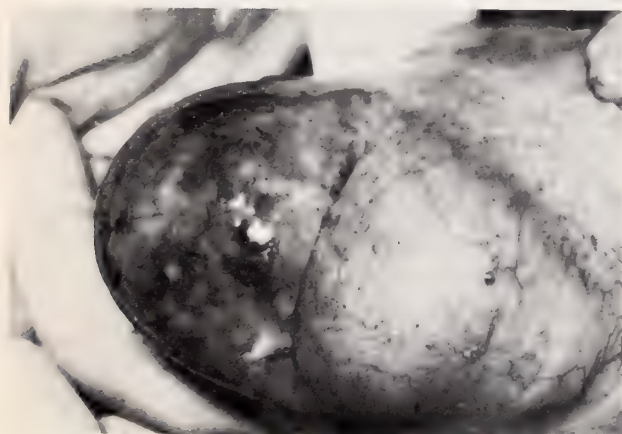


FIG. 1 Close-up photograph showing cecum as it was delivered from abdominal incision. The area of infarction is clearly visible on the left.

enema (5). Holmes reported mesenteric vascular occlusion in pregnant women that may be possibly related to dietary ergot toxicity (10). In 1947, Berman reported a case of massive intestinal infarction caused by mesenteric thrombosis in a patient taking large amounts of ergot (4). Microscopic examination was not reported. Green documented mesenteric vasospasm by arteriography in a middle-aged white woman who had peripheral vascular complications of ergotamine but no abdominal complaints (6). Katz reported abdominal angina related to ergot ingestion (11). Myocardial ischemia and related infarction have also been reported (7). One report suggests ergot-containing compounds may be the treatment of choice for opiate refractory diarrhea (12).

Although pathologic examination of the normal and necrotic bowel in our specimen does not aid in making a firm diagnosis of the etiology of the infarction, several processes can be eliminated. Cecal volvulus is excluded since the operative findings revealed no evidence of a redundant or elongated mesocolon and no proximal distention of the small bowel. There was no evidence of inguinal and/or femoral hernia preoperatively, intraoperatively or immediately postoperatively, at two months, and at ten months follow-up. There was no apparent posterior or anterior pelvic wall defect, and the cecum was easily delivered into the abdominal wound thereby making strangulation secondary to hernia incarceration unlikely.

There was no evidence of foreign body perforation. There were excellent pulsations present in all branches of the superior and inferior mesenteric vessels thereby eliminating a central mesenteric phenomenon. We postulate that vasoconstriction of the most distal branches of the superior mesenteric arterial system related to ergot ingestion was sufficient to produce transient intestinal ischemia and resultant infarction.

The possibility of hypotension related to Dilaudid injection as an accessory factor exists. Microscopic examination with a polarized microscope revealed no evidence of particular refractory matter, that is, injected debris related to the Dilaudid injection. One would have to postulate the existence of an arterial venous communication such as an arteriovenous fistula, or cardiac septal defect, none of which are present in our patient, in order to obtain passage of particular matter into the cecum. It is of interest that the infarcted area is in the region supplied by the ileocolic artery, the terminal extension of the superior mesenteric, an artery known to be readily accessible to blood-borne emboli. Glauser, in reviewing medical and surgical sequelae of

drug abuse and overdose, makes no mention of intestinal infarction other than that of cecal dilatation secondary to pseudo-obstruction which may necessitate colonic decompression (2). Although our patient did exhibit mild dilatation of the cecum, it was markedly less than the 12 or 13 cm dilatation that is usually associated with cecal infarction.

Although our patient's recent and past history of drug addiction and abuse could correlate well with his highly anxious and agitated state upon admission, it must be noted that ergot-containing compounds can produce a variety of mental disturbances. It is interesting to speculate whether or not some aspects of the psychiatric and social disturbances exhibited in our patient are related to ergot-containing compounds.

The above discussion suggests that there may exist a more than casual relationship between the ingestion of ergotamine and the intravenous injection of Dilaudid tablets and the development of cecal infarction in our patient. Although the role of ergotamine cannot be clearly documented as an etiologic factor in our case, a review of the literature suggests that ergot compounds may be capable of eliciting our patient's pathology. The medical community needs to be more cognizant of the adverse effects of ergot-containing compounds.

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Dermatologic Pseudobenignities

JOHN KWITTKEN, M.D.

The dermatologic malignancies, premalignancies, potential malignancies, and pseudomalignancies have been classified in a previous communication (1). Surprisingly, virtually nothing has been written about the dermatologic pseudobenignities. These are lesions that appear histologically benign to the unwary pathologist, but always show evidence of aggressive biologic behavior. It is exceedingly important to be able to recognize these lesions, if possible, so that they may be thoroughly eradicated and/or so that investigations may be performed that might uncover a malignant systemic disease.

This paper classifies the dermatologic pseudobenignities in terms of primary germ layer origin and the type of tissue involved in each germ layer (Table I) and briefly discusses the salient features of each pseudobenignity listed.

Basal Cell Carcinoma

There are two types of basal cell carcinomas which, to the untrained pathologist, frequently have been diagnosed as benign lesions. One form is the creeping basal cell carcinoma composed either of multiple small superficial buds or slender anastomosing cords of basalioma cells simulating either a flat or reticulated seborrheic keratosis, respectively. Features that help to define these lesions as malignancies include retraction artifact and stromal proliferation at the periphery of these neoplastic nests and cords, and the nature of the nuclear chromatin pattern (1, 2). In basal cell carcinomas, the chromatin particles are coarse and crowded together, presenting a stippled pattern. While in seborrheic keratoses, the nuclei of the basaloid cells have fine, evenly spaced, inconspicuous particles forming a lacy network. The presence of melanin is an addi-

tional aid. In seborrheic keratoses, the melanin granules are fine and golden and form a compact perinuclear cap. In basal cell carcinomas, the melanin granules are coarse, dark brown, anisocytotic and randomly distributed.

A sclerosing basal cell carcinoma containing many keratinized cells and keratin-filled ductular structures and composed primarily of slender anastomosing cords of basalioma cells may simulate a sebaceous duct adenoma (so-called syringoma) or a trichoepithelioma. The sebaceous duct adenoma can be distinguished by the presence of ductular structures with a luminal layer of vacuolated cells. Also, when keratin-filled ductular structures are present, the lining always shows epidermal keratinization (a granular layer is present).

Unfortunately, it is not always possible to distinguish the sclerosing basal cell carcinoma from a trichoepithelioma histologically. Not only may the trichoepithelioma show similar stromal changes (both sclerotic and loose proliferation with retraction artifact), but also the epithelial cell linings of the keratin-filled ductular structures in each may or may not have a granular cell layer. Even the nuclear chromatin pattern of the basalioma cells and the basaloid cells of the trichoepithelioma are identical. The presence, of course, of a lobular pattern, a lacelike network, abortive hair papillae and a large number of keratin-containing cystic structures with or without hair shafts favors a trichoepithelioma.

Keratocarcinoma

Although formerly termed keratoacanthoma and considered to be a premalignancy (3, 4), it is now becoming increasingly evident to many authorities that the keratocarcinoma (5) is a true primary cutaneous malignancy that progresses through characteristic stages during its development. The incipient stage lesion often is diagnosed as a benign lesion. The diagnoses most frequently made are verruca vulgaris, seborrheic keratosis, inverted follicular keratosis and lichen planus. Although his-

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TABLE I
Dermatologic Pseudobenignities

Ectodermal Origin
Epithelial Tissue
Basal cell carcinoma, creeping and sclerosing types
Keratocarcinoma, incipient stage
Mesodermal Origin
Connective Tissue
Dermatofibrosarcoma protuberans
Desmoid Tumor
Differentiated Liposarcoma
Infiltrating angiolipoma
Infiltrating lipoma
Malignant fibrous histiocytoma
Lymphoreticular Tissue
Cutaneous plasmacytoma
Leukemia cutis, chronic lymphocytic type
Lymphoma cutis, well differentiated lymphocytic and follicular types
Mycosis fungoides, erythematous stage (localized or generalized)
Parapsoriasis variegata or en plaques (some cases)
Poikiloderma (some cases)
Vascular Tissue
Kaposi's sarcoma, early and angiomatous lesions
Malignant hemangiopericytoma

tologically benign, the incipient stage lesion shows a combination of features that are unique. These consist of simultaneous increase in rate of cell production with epidermal hyperplasia and premature keratinization of epidermal cells. Although a *verruca vulgaris*, a seborrheic keratosis and lichen planus (especially the hypertrophic form) must be considered microscopically in the differential diagnosis, the absence of premature keratinization in the first two diseases and the presence of an inflammatory cell infiltrate associated with basal cell destruction in the last disease excludes them. The inverted follicular keratosis may be excluded by the conspicuous absence of squamous cell eddies. It is extremely important to recognize this stage and thoroughly eradicate such lesions to prevent future disfiguring scars and/or possibly destructive or fatal invasive and metastatic disease. Not generally appreciated, but of considerable prognostic importance, are the incipient stage epidermal changes extending to the excised margins of many persistent and recurring late stage lesions. In other words, the border of an actively growing lesion is not limited to but extends beyond the lip of the crateriform depression. This is probably the reason for the high recurrence rate of many surgically excised lesions.

Dermatofibrosarcoma Protuberans

The dermatofibrosarcoma protuberans (3) is a low-grade fibrosarcoma characterized generally by compact interlacing bundles of spindle-shaped cells

associated with a storiform pattern, varying degrees of nuclear hyperchromatism and pleomorphism, relatively frequent mitotic activity and attenuation of the overlying epidermis with focal ulceration. It is a locally invasive tumor and rarely metastasizes. There are occasional lesions, however, that mimic the benign cellular dermatofibroma (fibrous histiocytoma) histologically, and it is only with the passage of time that evidence of the aggressive biologic nature of these pseudobenignities becomes manifest. Because of this, it is recommended that patients having skin lesions diagnosed as a cellular dermatofibroma have frequent follow-up visits. At the first sign of a recurrence, adequate re-excision of the area should be performed.

Desmoid Tumor

The desmoid tumor is a locally aggressive fibromatosis (3, 6) that is considered to arise from a muscular aponeurosis. Histologically, it appears benign and is characterized by interlacing bundles of fibroblasts and focally hyalinized collagen bundles without nuclear atypism and showing rare mitotic activity. Foci of mucoid degeneration, calcification, microhemorrhage, hemosiderin deposition and inflammatory cell infiltrates are often encountered. Characteristically, this lesion invades voluntary muscle, often tendons and occasionally internal viscera. It frequently recurs locally after surgical excision. It is a potentially dangerous and sometimes fatal lesion. When first diagnosed, desmoid tumors should be treated by radical local excision including a wide margin of the uninvolved tissues surrounding the grossly visible lesion whenever it is surgically feasible.

Differentiated Liposarcoma

The differentiated liposarcoma (3, 7) is a low-grade sarcoma arising from mesodermal cells showing a differentiation into fat cells. It rarely arises in the subcutaneous fat and usually develops within the intermuscular fascial planes with extension to the subcutaneous tissue. It has a predilection for the thighs. Microscopically, this lesion is often composed primarily of mature fat cells and blood vessels showing varying numbers of fusiform and stellate cells, occasional lipoblasts and often a myxomatous stroma. It possesses no true capsule and often attains a large size. This lesion has a high rate of recurrence and a small percentage metastasize. Because of the paucity and occasional absence of bizarre lipoblasts, these lesions are often interpreted as benign. This true pseudobenignity often is diagnosed correctly as a malignancy retrospectively following clinical evidence of metastatic disease. It

is recommended that a wide excision should be performed on all fatty tumors which show even the slightest degree of cellular atypism and immaturity in order to avoid recurrent and/or metastatic disease.

Infiltrating Angiolipoma and Infiltrating Lipoma

The infiltrating angiolipoma and infiltrating lipoma are deep-seated, unencapsulated tumors, usually involve the subcutaneous tissue, and always invade skeletal muscle (8, 9). The former is composed primarily of a mixture of mature adipose tissue and blood vessels; the latter is composed mainly of mature adipose tissue. Atypia, pleomorphism and mitotic figures of both adipose and angiomatous components are never encountered. These are locally aggressive lesions that are difficult to eradicate, may grow to extremely large dimensions, may invade fascia, joint capsules, bones and nerves, and have a high recurrence rate following surgical excision. The explanation for the high recurrence rate is most likely because of inadequate excision, since the gross boundaries of these lesions are ill defined. Although histologically benign, these are true pseudobenignities because of their locally invasive and destructive growth qualities. In all cases, a wide excision should be performed to include normal surrounding tissue.

Malignant Fibrous Histiocytoma

The malignant fibrous histiocytoma (10), like dermatofibrosarcoma protuberans, may mimic, histologically, the benign cellular dermatofibroma (fibrous histiocytoma). The presence of giant cells and foamy histiocytes in variable numbers distinguishes this lesion from dermatofibrosarcoma protuberans. Criteria for malignancy include penetration of fascia, muscle and/or bone, angioinvasion and multinodular local growth. Since none of the above criteria may be present, it is recommended, as for the dermatofibrosarcoma protuberans, that all patients with lesions diagnosed as a cellular dermatofibroma be followed frequently with adequate re-excision at the first sign of recurrence.

Cutaneous Lymphoreticular Pseudobenignities

Perhaps the most frequently encountered group of cutaneous pseudobenignities involves diseases composed primarily of mononuclear cells of lymphoreticular origin. In most cases the malignant nature of these lesions cannot be unequivocally diagnosed from the histology alone; in fact, the microscopic features are indicative of benign lesions

in many instances. It is only with the help of associated clinical disease and/or the progression of these lesions with the passage of time that the diagnosis of a definite malignancy can be made.

The cutaneous plasmacytoma, a tumor derived from B lymphocytes, is rarely a primary lesion (11,12). It is most often secondary to coexisting multiple myeloma. It may represent either a metastasis or direct extension from underlying bone disease. In a small percentage of cases, these lesions may herald the full-blown clinical disease by several years. Although in many instances the histologic features of the neoplastic cells are sufficiently atypical to warrant a diagnosis of plasmacytoma, there are also cases in which the cellular infiltrate cannot be distinguished from normal mature plasma cells such as those that occur in conspicuous tumorous aggregates in several infectious and noninfectious diseases. With coexisting multiple myeloma, the diagnosis is simple. In its absence, it is wiser to label these lesions as cutaneous plasmacytic tumor, type and biologic nature undetermined. These patients should be thoroughly investigated for the presence or absence of multiple myeloma and other nonneoplastic inflammatory diseases (including syphilis) associated with tumorous plasma cell infiltrates.

Probably the most frequently encountered pseudobenignity is the presence of spotty, often perivascular, infiltrates of mature-appearing lymphocytes involving various levels of the cutis occurring in patients which either have or are later proven to have chronic lymphocytic leukemia or a well differentiated lymphocytic lymphoma. Histologically, these lesions cannot be distinguished from other benign inflammatory conditions associated with conspicuous lymphocytic infiltrates. These findings, in the absence of coexisting leukemic or lymphomatous disease, should be interpreted with caution by the examining pathologist. I would suggest that these lesions be diagnosed lymphoreticular proliferation, type and biologic potential undetermined, and that such patients be thoroughly investigated for the presence or absence of systemic lymphoreticular disease. Fortunately, rarely encountered but also creating a similar diagnostic problem and investigative procedure, is the cutaneous lymphoma showing a well defined follicular or nodular pattern (13) simulating lymphocytoma cutis and other benign inflammatory disorders.

Some cases of parapsoriasis (variegata or en plaques) and poikiloderma, which finally eventuate into classic mycosis fungoides or frank lymphoma, represent true pseudobenignities either *ab initio* or developing from parapsoriasis or poikiloderma, appearing as benign inflammatory conditions histolog-

ically. Similarly, in most instances the erythematous stage (localized or generalized erythrodermic form) of mycosis fungoides cannot be diagnosed initially by histology alone. The presence of scattered atypical mononuclear cells in the inflammatory cell infiltrate does not confirm the diagnosis since similar cells may be seen in clearly benign inflammatory processes. The diagnosis is often made retrospectively when the patient later progresses to the characteristic plaque and/or tumor stage. In all likelihood, because of our relatively unsophisticated techniques with light microscopy, the early lymphomatous cellular components cannot be detected. Continuing along the same vein, some authorities consider these conditions to represent "premycotic" lesions, a concept which I cannot accept. A lesion is either benign or malignant. To postulate the existence of an "in-between" stage is not only untenable and unscientific, but also seems to represent an attempt to hide our human diagnostic failings.

Kaposi's Sarcoma

Kaposi's sarcoma is a malignant lesion, most often involving the skin of the lower extremities, presumably derived from immature endothelial cells and pericytes which are considered responsible for the angiomatous and fibroblastic components, respectively. Although in most instances a histologic diagnosis is easily made, in occasional early and predominantly angiomatous lesions showing virtually no endothelial atypism, vascular slits or fibroblastic activity, a diagnosis is not possible on histologic grounds alone. The microscopic features are often indistinguishable from stasis dermatitis which, incidentally, often coexists. The correct diagnosis of this pseudobignity is only reached because of the presence of other characteristic lesions and/or progression of the lesion.

Malignant Hemangiopericytoma

The hemangiopericytoma is considered to be a vascular tumor derived from pericytes (14). It is characterized histologically by compact aggregates of small cells with scanty amounts of poorly defined eosinophilic cytoplasm and generally oval or spindle-shaped often hyperchromatic nuclei localized about small vascular channels. The presence of nuclear pleomorphism, prominent mitotic activity, increased cellularity, focal necrosis and hemorrhage are features indicative of malignancy. Lesions may be either entirely benign or malignant. Unfortunately, lesions that appear histologically benign have proven to be malignant clinically. It is there-

fore recommended that whenever a diagnosis of benign hemangiopericytoma is made, this lesion be thoroughly and widely excised, since what seems to be a benignity microscopically might actually represent a pseudobignity.

Summary

Dermatologic pseudobignities are cutaneous lesions that appear histologically benign to the unwary pathologist, but which always show evidence of aggressive biologic behavior. In most instances, a careful detailed analysis of both cellular detail and overall structure of the lesion will enable the pathologist to appreciate the aggressive or potentially aggressive nature of these lesions. In some instances, the aggressive nature can only be appreciated retrospectively. I have categorized and discussed their salient features. It is extremely important to recognize these lesions whenever possible, so that they may be thoroughly eradicated and/or that investigations may be performed which might uncover a malignant systemic disease.

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Neurofibromatosis of von Recklinghausen: A Rare Cause of Renovascular Hypertension

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Abstract

Neurofibromatosis of von Recklinghausen is an extremely rare cause of unilateral renovascular hypertension. A patient afflicted with this disorder and her dramatic response to nephrectomy is reported.

Hypertension associated with neurofibromatosis of von Recklinghausen may be caused by pheochromocytoma or by characteristic lesions involving the renal arteries. Pheochromocytoma is the usual cause of hypertension in the adult with this disease, while renal artery stenosis is much more common in children so afflicted. The renovascular lesions are usually diffuse and focal, most often bilateral, and involve the main renal arteries, the intralobar arteries, and arterioles. The narrowed vascular lumen results from disruption of the vessel wall by micronodules composed of proliferating cells, probably derived from axonal elements. Fibrosis and aneurysm formation are also frequently found. The renovascular lesions are progressive, as are most other lesions associated with neurofibromatosis, which explains the relatively poor results in achieving permanent amelioration of hypertension from renovascular reconstructive surgery and nephrectomy.

Case Report

R. M. was found to be severely hypertensive at age sixteen. Her father and paternal grandmother had von Recklinghausen's disease. At age twenty-two, a right hemicolectomy was performed for massive hemorrhage from polyps and hemangiomas. Diffuse angiomas involved most of the colon

and terminal ileum. The adrenal glands were normal, but the left kidney was smaller than the right. The blood pressure was 280/160 mm Hg. The blood urea nitrogen, creatinine, urinalysis, and urinary catecholamine values were normal. The regitine® test was negative and an intravenous pyelogram was unremarkable. Renal angiography was technically unsuccessful. Treatment with a variety of antihypertensive medication was of little benefit.

She became pregnant at age twenty-six. The blood pressure was 280/130 mm Hg. She had classical neurofibromatosis manifested by cafe-au-lait spots and cutaneous neurofibromatoses. Grade II hypertensive retinopathy was present, and significant left ventricular hypertrophy and enlargement were apparent on physical examination, electrocardiogram and x-ray film of the chest.

Following therapeutic abortion, renal angiography disclosed a small left kidney with an arterial supply comprised of three or four threadlike vessels originating from the aorta, and several collateral vessels arising from the adrenal circulation. The right renal artery and kidney appeared completely normal. Because of the severity and intractability of the patient's hypertension, a left nephrectomy was performed, following which, her blood pressure fell to 125/75 mm Hg. The left renal artery was almost totally obliterated by a process characteristic of von Recklinghausen's disease. One year later, the blood pressure was 165/95 mm Hg. without treatment and fell to normal with small doses of antihypertensive medication. Unfortunately, her subsequent course is unknown.

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Von Recklinghausen's disease associated with renovascular hypertension is quite rare, fewer than fifty cases having been reported. Medical therapy is preferable to surgery in the majority of patients because renal involvement is usually bilateral, nephrectomy must often be performed in lieu of vascular reconstruction, and the vascular lesions of neurofibromatosis are often progressive. In the un-

usual instance of unilateral renal involvement when there is a very substantial anatomical and functional difference between the two kidneys, an aggressive surgical approach may be rewarded with a dramatic result in patients whose hypertension is resistant to adequate medical therapy.

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Familial Cutaneous Leiomyomata: A Case Report with Electron Microscopic Study

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Abstract

A case of familial cutaneous leiomyomata is reported. The patient had multiple cutaneous nodules and cafe-au-lait spots, clinically misdiagnosed as neurofibromatosis. The histologic features of the painful and nonpainful tumors were compared and the ultrastructure of the nonpainful tumor was studied. In conjunction with the report in the literature, our findings could support the idea that compression and damage of the nerve fibers by the growing tumor may cause pain.

Introduction

Cutaneous leiomyomata are uncommon benign tumors of the smooth muscle in the skin. Except for their painful nature and possible familial occurrence, they present no specific clinical features and are usually misdiagnosed clinically (1). In contrast, the histology is usually simple and poses no problem in interpretation. This manuscript illustrates such an example.

Case Report

J.A. is a 40-year-old Caucasian female who was admitted for removal of a painful nodule on the right arm. The patient first noted the lesion at puberty with subsequent appearance of many similar lesions irregularly distributed and found mainly on the trunk and upper extremities. A few were also seen on the legs. They were generally asymptomatic, except for the right arm nodule, which had become painful when it increased in size and had been removed previously because of pain. All lesions were raised, smooth, reddish-brown, firm nodules measuring approximately 1 cm (Fig. 1).

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Concomitant with these nodules were a few irregular, flat pigmented areas in the skin. On the back, was a brown macule present since birth, and on the right wrist was a 3 cm × 4 cm brown macule (Fig. 2) that first appeared at puberty. Because of the cafe-au-lait spots and multiple cutaneous nodules, the diagnosis of neurofibromatosis was entertained clinically.

The family history revealed that several members from three generations of her family had similar multiple cutaneous nodules (Fig. 3). At 26 years of age, the patient underwent a hysterectomy for a cellular uterine leiomyoma that had few bizarre giant cells and rare mitosis. Her mother also had a hysterectomy for a uterine leiomyoma.

Materials and Methods

The painful nodule from the patient's right arm was excised. A nonpainful nodule from her back was later biopsied for comparison. A nodule from her mother was also biopsied for diagnosis.

All specimens were fixed in 10% formalin and processed in routine fashion, stained with hematoxylin and eosin. Fresh tissue from the nonpainful nodule was available for electron microscopic study after it was fixed in buffered 4% formaldehyde and 1% glutaraldehyde, processed in conventional ways and stained with uranyl acetate and lead citrate.



FIG. 1 Extensor surface of the upper arm showing multiple raised, dark nodules.



FIG. 2 A large café-au-lait spot on the forearm.

Results

Grossly, the skin biopsy from the painful lesion of the right arm revealed a 6 mm × 6 mm tan, rubbery-firm nodule that, histologically, was an uncapsulated, somewhat circumscribed lesion involving the entire depth of the dermis (Fig. 4). It was composed of interlacing bundles of spindle cells with slender, blunt-end nuclei and a moderate amount of eosinophilic cytoplasm. The lesion was very cellular and compact, and no nerve could be recognized within the tumor. At the periphery of the tumor, smooth muscle cells surrounded but did not infiltrate the nerve (Fig. 5).

The nonpainful nodules from the patient and her mother grossly resembled the painful nodule as described above. Histologically, the two nonpainful nodules were indistinguishable. Both were ill-de-

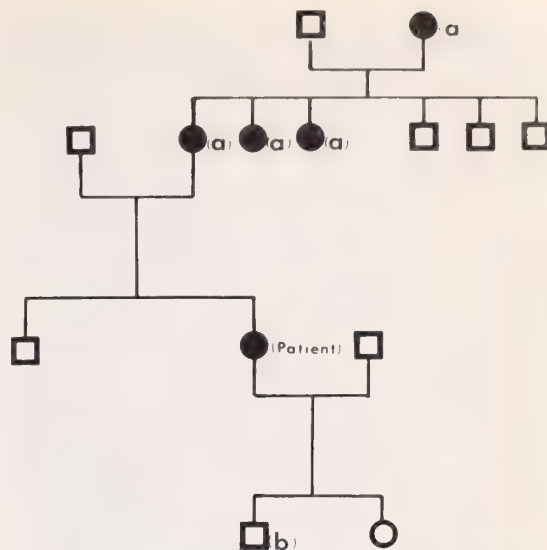


FIG. 3 (a) The affected females in the three generations of the patient's family have multiple cutaneous nodules similar to those present in the patient. (b) Her 14-year-old son has a large café-au-lait spot since birth.

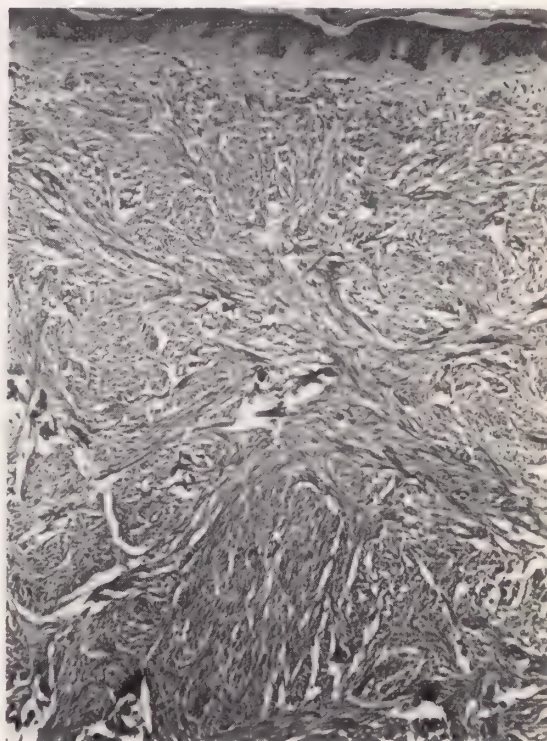


FIG. 4 Photomicrograph of the painful nodule showing the uncapsulated, rather circumscribed lesion which is very cellular, composed of fascicles of spindle cells. (Hematoxylin and eosin, ×48).

fined lesions, merging imperceptibly with adjacent dermis. They were much less cellular than the painful lesion and the smooth muscle cells proliferate loosely in between the dermal collagen fibers

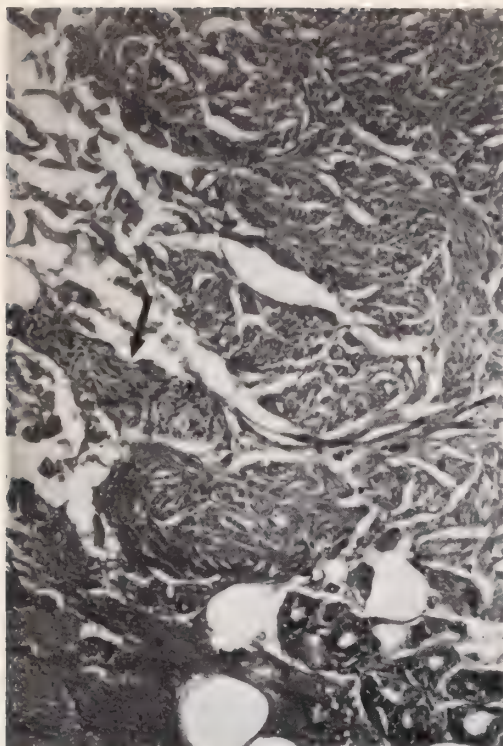


FIG. 5 A nerve (arrow) is surrounded by the smooth muscle cell from the painful tumor at the deep dermis. (Hematoxylin and eosin, $\times 48$).



FIG. 6 The nonpainful nodule is an ill-defined, less cellular lesion with proliferating smooth cells merging imperceptibly with dermal collagen. (Hematoxylin and eosin, $\times 48$).

which could still be well recognized within the tumor (Fig. 6).

The transmission electron microscopic study of the nonpainful nodule revealed a single spindle cell or groups of spindle cells being separated by collagen fibers. An individual spindle cell was encircled by a layer of basal lamina. The oval nucleus was central. The plasmalemma was studded with numerous pinocytic vesicles. Scattered in the cytoplasm were a few mitochondria, profiles of endoplasmic reticulum, free ribosomes, and abundant myofilaments that often aggregated into bundles of various densities with frequent formation of oval to fusiform dense bodies (Fig. 7). Loosely separated from the muscle cells by fibroblasts and collagen fibers was a mixed nerve containing both unmyelinated fiber and myelinated fiber with Schwann cells (Fig. 8). An intact layer of connective tissue cell enclosed the nerve fibers and the intervening collagen as a complete perineural sheath.

Discussion

The association of multiple cutaneous nodules and cafe-au-lait spots and a positive family history

certainly makes neurofibromatosis a very likely clinical diagnosis. The usual histologic feature of neurofibroma is that of trabeculated fascicles of spindle cells and may simulate a leiomyoma on some occasions (2). The histologic diagnosis of leiomyoma is usually easy and, in this case, the diagnosis was confirmed by ultrastructural study. It is noteworthy that isolated cafe-au-lait spots can be seen in 10% of the general population (2) and its association with leiomyoma in this patient was a chance occurrence.

Cutaneous leiomyoma may arise from arrector pilorum muscle in the dermis or from smooth muscle of mammary gland, scrotum, labia majora, and the wall of blood vessels (angioleiomyoma). Multiple cutaneous leiomyomata of pilar origin have been reported to affect identical twins (3) siblings (4,5) and three or four generations in the family. It was interesting that the affected members of the family reported here were all females. This, however, does not contradict the belief that this disease is inherited as an autosomal dominant disorder with incomplete penetrance (6).

The lesions usually appear around the age of 20 years, and occur in the trunk and extremities, more often in the extensor surface. Pain is a characteristic

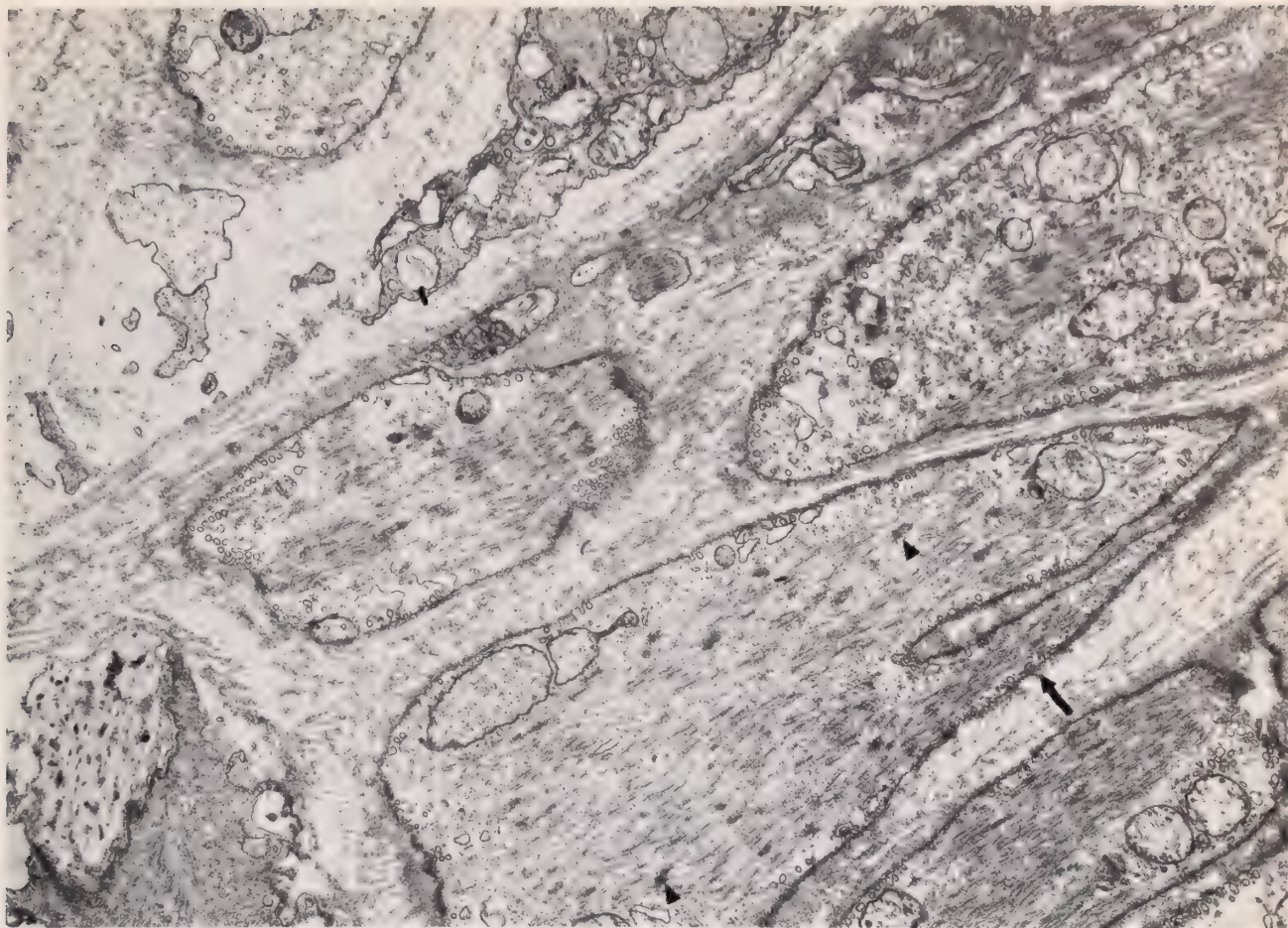


FIG. 7 Electron micrograph of the nonpainful tumor illustrating characteristics of smooth muscle cells: complete basal lamina (arrow) encircling individual cells, many pinocytotic vesicles along the plasma membrane, numerous myofilaments with dense bodies (arrow-head). ($\times 10,000$)

feature of these tumors, but its etiology is not known. Fisher and Helwig (1) were unable to correlate the painful symptoms with any specific lesion. However, Montgomery and Winkelmann (7) found that only the larger lesion that had invaded or extended around the nerve in the deep corium or subcutis by the tumor were painful. In our study, the very high cellularity, the compact and expansile pattern of growth, and the encroachment of a nerve by the tumor at the periphery were the histologic features that distinguished the painful nodule from the nonpainful lesion. No appropriate tissue from the painful tumor was available for electron microscopic study to allow ultrastructural comparison. However, our ultrastructural finding of a normal, unaltered nerve within the tumor was in contrast to the distorted and disrupted nerve and the absence of intact perineural sheath around the nerve

demonstrated by Mann (8) in his study of the painful cutaneous leiomyoma. Moreover, the exaggerated interdigitation in the compressed muscle cells described by Mann was not observed in this nonpainful tumor. In conjunction with Mann's finding, our observation might support the idea that compression of the nerve fibers by the larger tumor may damage the nerve and cause pain.

The association of cutaneous leiomyomata and uterine leiomyoma, as seen in this patient and her mother, has been previously reported (3,9) and there has been controversy as to its significance. While some authors (6) considered the connection not fortuitous, the authors of this paper agreed with Jolliffe (4) that the association could easily occur by chance since, in general population, uterine leiomyoma can be found in one out of four women in their reproductive life (10).



FIG. 8 Oblique section of a mixed nerve within the nonpainful tumor containing a myelinated nerve fiber (m), nonmyelinated nerve fibers (nm) and schwann cells (s). A well-preserved layer of connective tissue cell encloses the nerve fibers, collagen fibers (c) and forms the perineural sheath (p). ($\times 7,500$)

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Choledochal Cyst: Case Report and Review of the Literature

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Abstract

A 14-year-old female with a large choledochal cyst is presented. The radiologic work-up and operative findings of this clinically unsuspected abnormality are described and the entity of choledochal cyst is discussed.

Case Report

A 14-year-old girl was admitted to The Mount Sinai Hospital on May 2, 1979 with upper abdominal mass, intermittent abdominal pain and intermittent jaundice. The patient was in good health until the age of five, when she began complaining of intermittent abdominal pain. An upper gastrointestinal series and a barium enema at that time were reportedly normal. The pain spontaneously resolved and she was well until July 1978 when she began to have irregular menses. She was treated with Provera with some improvement. In December 1978, she noticed cessation of menses, increased fatigue, dark urine, light stools, pruritus, anorexia, and diarrhea. Abnormal laboratory findings at that time included: bilirubin 9 mgr, SGOT 350 and alkaline phosphatase 1100 MU/ml. A tentative diagnosis of cholestatic hepatitis was made. Her symptoms persisted and, in February 1979, a liver biopsy showed cholestasis, chronic inflammation and early periportal fibrosis. The symptoms of jaundice and fatigue began to resolve in March but they recurred in April. The repeat bilirubin was 1.7 mgr, SGOT 25, SGPT 109, and alkaline phosphatase 1000 MU/ml. At that time the patient's mother noticed her

increasing abdominal girth. An upper gastrointestinal series revealed a large mass compressing the second, third and fourth portions of the duodenum (Fig. 1).

On admission, the patient was icteric. There was a large nontender, nonmobile mass in the right upper quadrant extending to the left of the midline. The mass was smooth in contour and distinct from the smooth liver edge.

The admission laboratory data revealed a bilirubin of 3.5 mgr, SGOT 177, SGPT 162 and alkaline phosphatase of 990 MU/ml. The hemoglobin was 13.7, hematocrit 39.4, white blood count 7,500. Prothrombin time and fibrinogen were within normal limits. The chest x-ray was normal.

The patient had an abdominal sonogram that revealed a large cystic structure 13 cm in size in the region of the common bile duct. A normal sized gallbladder was also visualized (Fig. 2). On May 8, 1979, the patient underwent a percutaneous transhepatic cholangiogram, using the fine "chiba" needle (outer diameter 0.7 mm) that revealed moderate dilatation of the distal intrahepatic ducts (Fig. 3). The gallbladder and the cystic duct were slightly dilated. The entire common bile duct was replaced by a giant cystic dilatation. There was no passage of contrast material into the duodenum noted.

Following the transhepatic cholangiogram, the patient underwent exploratory laparotomy. A large (16 cm in diameter) choledochal cyst was found at exploration that contained approximately 1000 cc of bile and sludge. The cyst wall was fibrotic, 3 to 4 mm in thickness. It extended from just above the

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FIG. 1 Upper gastrointestinal examination. Obvious widening and compression of the duodenal loop and distal antrum of the stomach by a mass lesion.

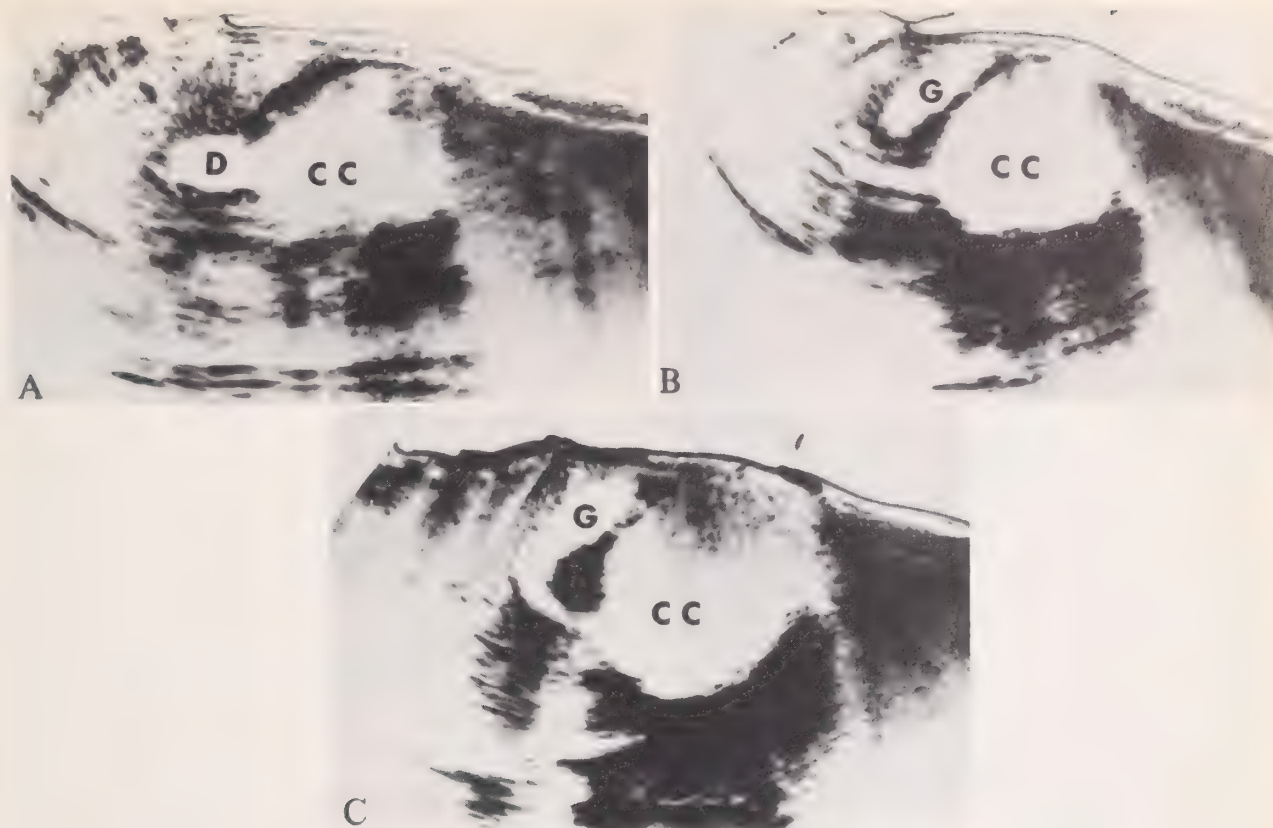


FIG. 2 Longitudinal views (A,B,C) from an abdominal sonogram show the large choledochal cyst (CC) and dilated ductal system (D). A relatively normal sized gallbladder (G) is seen in views B and C.

bifurcation of the hepatic duct to include the entire length of the visible common duct. The right and left hepatic ducts were significantly dilated—measuring 2.5 and 3 cm respectively—and each entered the cyst separately. The gallbladder appeared to be normal. The cystic duct was dilated to approximately 1 cm and entered the cyst separately, approximately 1 cm below the hepatic ducts (Fig. 4). The distal opening of the cyst was not identified. The pancreas and duodenum were splayed out over the anterior surface of the cyst. The pancreatic duct was not identified.

The patient underwent a cholecystectomy and a side-to-side roux-en-y cysto-jejunostomy. Final pathology report revealed a normal gallbladder. The wall of the choledochal cyst showed partial destruction of the epithelial lining, mild chronic inflammation and fibrosis. Needle biopsy of the liver at the time of surgery revealed severe portal fibrosis and ductal proliferation compatible with obstructive biliary cirrhosis. The postoperative course was benign. The bilirubin dropped to 1.1 mgr. SGOT to 64, SGPT to 60 and alkaline phosphatase to 359.

She was discharged on the twelfth postoperative day. Four months postoperatively, the patient was asymptomatic and no mass was palpable.

Discussion

Choledochal cyst is an uncommon abnormality of the biliary tree. It has been seen in all age groups, although the majority of patients are children and young adults. It has been said to be four times more common in females (1) than in males, and most cases are in the first two decades of life. Several attempts have been made at classification of the various types of cystic dilatation of the biliary ductal system (2,3). The categories range from single cyst to multiple cysts of the intrahepatic ducts without abnormalities of the extrahepatic biliary system to marked dilatation of the latter as described in this report.

The classical triad of abdominal pain, mass and jaundice is not always present and results of blood chemistries may vary with the stage of the disease.

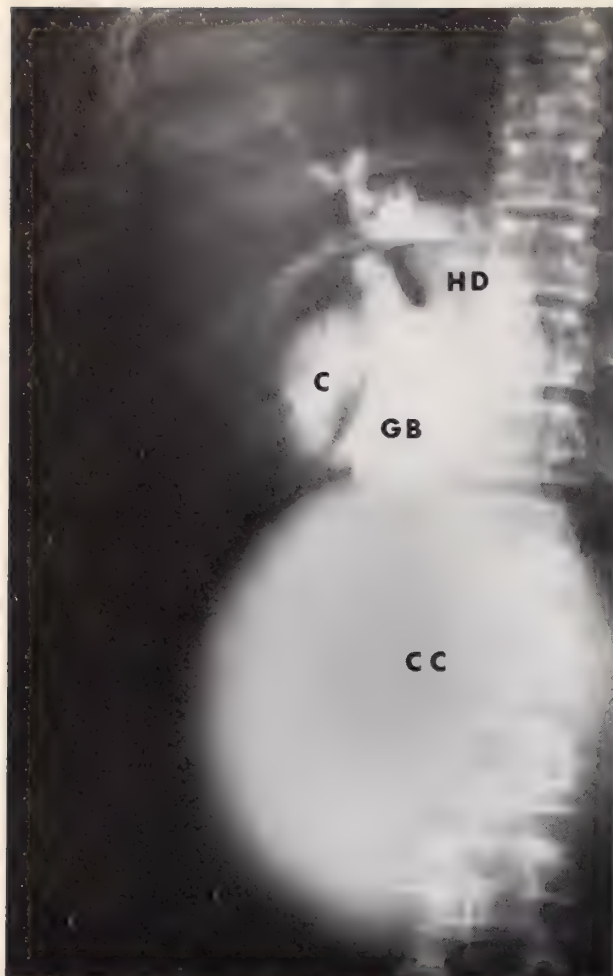


FIG. 3 Transhepatic cholangiogram. After 200 cc of contrast material (Rinografin 60%) were injected via a puncture of the right hepatic duct, the patient was positioned supine to allow contrast material to reflux into the ductal system proximal to the puncture site.

CC = Choledochal cysts GB = Gallbladder C = Cystic duct HD = Right hepatic duct.

Because the symptoms of the choledochal cyst are often nonspecific and intermittent, they may be attributed to other, more common, entities and diagnosis may be delayed. Progressive bile stasis can result in liver failure and, therefore, early diagnosis is important.

Many theories have been proposed regarding the etiology of choledochal cyst (4-9). Most of these attribute at least part of the cause to congenital abnormality. One of the theories that is widely accepted (2) is based on an anomalous arrangement of the pancreaticobiliary ductal system secondary to failure of the normal separation of the pancreatic duct and common bile duct during embryologic development. Normally, the common bile duct and pancreatic duct are a common channel for a distance of 5 mm or less. In the anomalous situation,

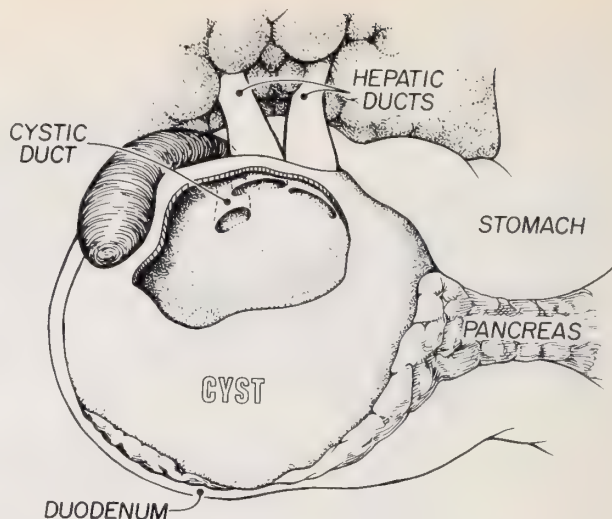


FIG. 4 Schematic drawing showing the anatomical situation of the ductal system at surgery.

the distal common bile duct enters the duct of Wirsung at a right angle approximately 2-3.5 cm from the ampulla of Vater. Since this anomaly precludes the development of a sphincter at the junction of the pancreas and common bile duct, pancreatic juice refluxes into the common bile duct. This results in recurrent bouts of cholangitis and dilatation of the common bile duct.

Treatment is surgical, usually cystojejunostomy or excision of the cyst.

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Clear Cell Acanthoma: A Metabolic Variant of Seborrheic Keratosis

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For several years, I have noted on a number of occasions a skin lesion showing the histologic features of both a seborrheic keratosis and a clear cell acanthoma. To the best of my knowledge, this observation has not been previously reported. During a recent period of four consecutive months, I collected five such cases that represent the subject of this report.

Materials and Methods

Five lesions were biopsied from five patients and were fixed in Bouin's solution, routinely processed, embedded in paraffin, sectioned, and stained with hematoxylin and eosin and the periodic acid-Schiff reaction (PAS) with and without diastase digestion.

The clinical data are summarized in Table I. All lesions were indurated, elevated, dome-shaped tumors with both scaly and smooth surfaces. Case 1 was brown; cases 2 and 3 were erythematous; case 4 was reddish-tan; case 5 was skin-colored. All lesions were removed by surgical excision with cauterization of the base and there have been no recurrences.

Histologically, all lesions showed the characteristic features of both an acanthotic seborrheic keratosis and a clear cell acanthoma in varying proportions. The clear cell acanthoma portion represented approximately fifty, twenty-five, ninety, fifty and seventy-five percent of the lesion in cases 1 through 5, respectively. There was an abrupt transition at the junction of both portions (Fig. 1). In cases 4 and 5, two transition zones were present, since the clear cell acanthoma portion occupied the center of each lesion. The portion having the fea-

tures of a seborrheic keratosis showed a mildly papillomatous and focally hyperkeratotic acanthotic epidermis composed mainly of lightly glycogenated uniform basaloid cells that contained large amounts of melanin in case 1 (Fig. 2). Horn cysts were present. The portion having the features of a clear cell acanthoma showed a focally crusted acanthotic epidermis, assuming pseudocarcinomatous proportions in case 5, diffuse parakeratosis, and absence of the granular layer. Except for the cells comprising the acrosyringium, walls of hair follicles and basal layer, virtually all of the remaining keratinocytes were enlarged and generally polygonal with normal-appearing nuclei, abundant amounts of glycogen-rich cytoplasm presenting a focally vacuolated, pale eosinophilic appearance with hematoxylin and eosin and prominent well-preserved intercellular bridges (Fig. 3). No melanin could be demonstrated within these cells in all cases. Its absence in this portion created an even sharper contrast to the portion showing the features of a pigmented seborrheic keratosis in case 1.

The subepidermal zone in the portion having the features of a clear cell acanthoma showed conspicuous vascular ectasia associated with edema and perivascular infiltrates of lymphocytes, histiocytes, neutrophils and melanophages. Foci of exocytosis associated with spongiosis of the epidermis were prominent in all cases. The subepidermal zone in the portion having the features of a seborrheic keratosis showed similar inflammatory changes except for minimal vascular ectasia. Also, foci of exocytosis associated with spongiosis of the epidermis were generally slight.

Discussion

The cases under discussion showed the characteristic histopathology (1) of both a seborrheic keratosis and a clear cell acanthoma occurring together

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TABLE I
Clear Cell Acanthoma: A Metabolic Variant of Seborrheic Keratosis

Case	Age	Sex	Location	Size*	Clinical Impression
1	57	M	Chest	0.8 cm	Intradermal nevus
2	75	M	Abdomen	1.0 cm	Seborrheic keratosis
3	73	M	Leg	0.7 cm	Seborrheic keratosis
4	60	M	Abdomen	1.2 cm	Seborrheic keratosis
5	72	F	Leg	1.5 cm	Basal cell carcinoma

* Size represents maximum dimension.

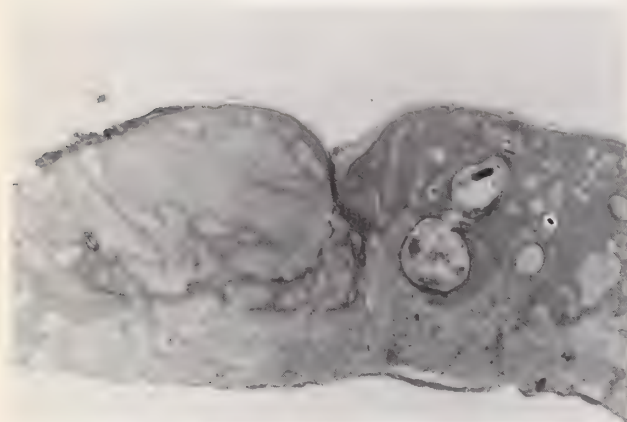


FIG. 1. Scanning view of Case 1 showing junction of seborrheic keratosis with clear cell acanthoma portion (H & E $\times$ 20).

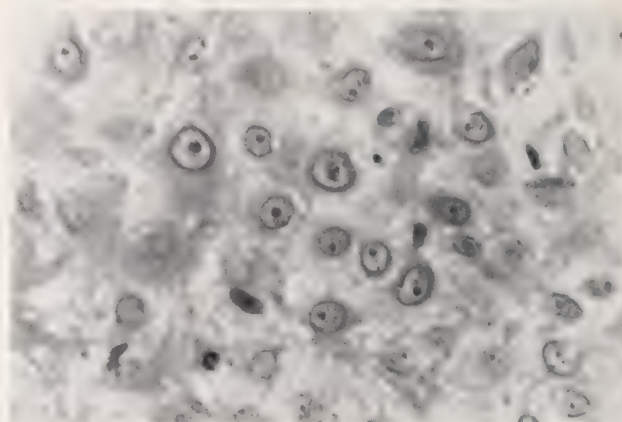


FIG. 3. High magnification of Case 1 detailing large glycogen-rich cells devoid of melanin of clear cell acanthoma portion (H & E $\times$ 400).

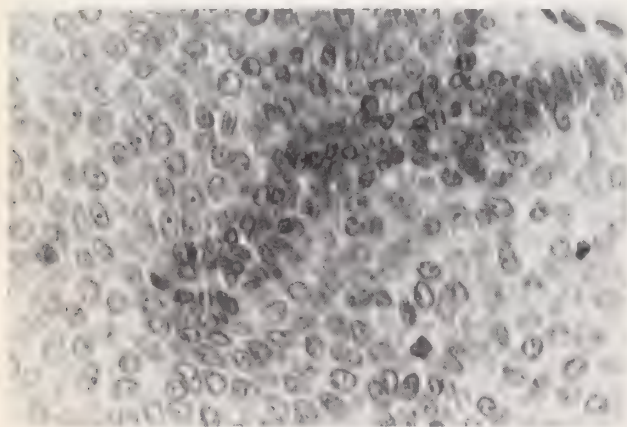


FIG. 2. High magnification of Case 1 demonstrating uniform basaloid cells of seborrheic keratosis portion. Dark band in center represents melanin-containing basaloid cells. (H & E $\times$ 400).

in each lesion. This association immediately raises some interesting questions. Is one dealing with one lesion or two different lesions located side by side? Could it be possible that the clear cell acanthoma is actually a seborrheic keratosis that has gone metabolically "haywire?" The answer to the first question is obvious. It must be one lesion. Not only is there epidermal continuity in all lesions but, in cases 4 and 5, the clear cell acanthoma portion was located in the center. As implied by the second

question, I believe that the clear cell acanthoma is a seborrheic keratosis which, by our relatively unsophisticated methods with light microscopy, seems to be composed largely of keratinocytes that do not have the ability to receive melanin and that accumulate large amounts of cytoplasmic glycogen because of either increased production and/or decreased or absent glycolysis. These changes seem to represent an unusual response to irritation, since all of these lesions are actively inflamed. Since the majority of them occur in the elderly, particularly on the lower extremities, an associated element of vascular insufficiency might possibly play a causative role.

It should also be noted that, while only trace amounts of glycogen can be demonstrated in keratinocytes of normal adult epidermis with the PAS technique, goodly amounts are easily demonstrated in keratinocytes of fetal epidermis, various nonspecific inflammatory reactions often associated with epidermal hyperplasia, viral infections such as the verruca vulgaris and molluscum contagiosum, lichen planus, psoriasis and various skin tumors including the seborrheic keratosis, eccrine poroma, basal cell carcinoma and squamous cell carcinoma. However, the quantity of glycogen observed in these conditions never approaches the exorbitant

amounts that are demonstrated in the clear cell acanthoma.

Thus, it appears that one is dealing with an irritated seborrheic keratosis showing focally the typical histologic features of a clear cell acanthoma. It seems most likely that the clear cell acanthoma actually represents almost total transformation of an irritated seborrheic keratosis beyond normal recognition. In effect, therefore, both the lesions under discussion and the clear cell acanthoma are irritated seborrheic keratoses showing yet another histologic alteration in the ever changing seborrheic keratosis (2). This lesion, while uncommon, does not seem to be rare, since it represents approximately 0.5 percent of all seborrheic keratoses examined in my laboratory during a period of four consecutive months.

Summary

Five skin lesions showing the combined histologic features of a seborrheic keratosis and a clear cell

acanthoma are reported, representing approximately 0.5 percent of all seborrheic keratoses examined. It is suggested that the clear cell acanthoma actually represents an irritated seborrheic keratosis beyond normal recognition because of almost total metabolic transformation of its nonadnexal epidermal keratinocytes (excluding the basal layer) into large glycogen-rich cells devoid of melanin. Both the lesions under discussion and the clear cell acanthoma can be added to the list of histologic alterations occurring in a seborrheic keratosis.

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Seminars in Reproductive Medicine

Forward

Seminars of Reproductive Medicine will be a new section in The Mount Sinai Journal of Medicine. It will present the best of the weekly departmental staff conferences and will offer a forum for a broad review which has become increasingly difficult to obtain from specialty journals.

Norbert Gleicher, M.D., Editor

Management of Anovulation

J. VICTOR REYNIK, M.D.

Most definitions pertaining to the disturbances of the normal female reproductive cycle are purely descriptive and simplistic. Neither do they imply etiologic factors nor are they quantitative. Thus, "anovulation" may be seen in a variety of clinical pictures, such as primary or secondary amenorrhea, oligomenorrhea, or dysfunctional uterine bleeding—symptomatic gradation not necessarily occurring in any sequence. They may be accompanied by symptoms of defeminization.

The normal female reproductive cycle is a dynamic system, characterized by constant fluctuations that are long-term (monthly) and short-term (minute to minute). Once those fluctuations are abolished, for whatever reason, the ovulatory process ceases.

Prior to discussing the clinical evaluation and therapy of anovulatory states, we will review the recent progress in neuroendocrinology, and the key points of the physiologic, ovulatory cycle.

Our understanding of the mechanisms regulating the ovulatory menstrual cycle has been significantly affected by the progress in neurophysiology. Most

importantly, it has been accepted that the brain acts as the master endocrine organ, which affects and controls the interacting elements of the reproductive axis: the hypothalamus, the pituitary and the ovary. Within this axis, the ovary seems to play a secondary role, consisting of providing the feedback signal to the brain which, in turn, controls the release of pituitary gonadotropins. By this feedback signal, the ovary does, however, ultimately determine the timing of ovulation. Within the framework of this concept, it is only logical to conclude that the etiologic basis for most ovulatory disturbances is most frequently of central origin. The main ovarian feedback signal to the central nervous system integrating unit is estradiol. While most of this steroid is of ovarian origin, only one-third of estrone is produced here; the remaining two-thirds are derived from the extragonadal conversion of androstenedione, as well as the metabolism of estradiol (1).

Ovarian production of progesterone reaches the maximum in about eight days after the LH surge (2). The life span of the corpus luteum in the primate seems to be constant and preprogrammed for about 14 days. Although the precise factors involved in the demise of this mini-endocrine organ are yet to be found, both estrogens and prostaglandins produced by the corpus luteum appear to be luteolytic and may serve a self-destruct mechanism (3).

Recent research developments allow for greater insight into the regulation of the endocrine com-

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partments of the ovary. FSH causes proliferation of the granulosa cells and induces within them the appearance of aromatizing enzyme, which converts androgens to estradiol (4, 5). Further growth of the follicle is affected by synergistic action of FSH with estrogens (6, 7). Estrogens cause an increase in the number of FSH receptors and, subsequently, the LH receptors (8).

The result of LH action is the synthesis of androgens by the theca cells (9). Androgens diffuse into the follicular fluid where they are aromatized to estrogens by the granulosa cells. Via the capillary network around the follicle, estrogens reach the systemic circulation and are delivered to the target sites, that include the central nervous system (CNS)-hypothalamuspituitary circuit. In response, release of GnRH occurs, affecting the synthesis and release of pituitary gonadotropins. Estradiol also appears to act directly on the gonadotropes, causing the preferential release of LH at midcycle (10, 11).

The decrease in FSH during the late follicular phase may be a result of an inhibitory effect of estrogen upon the pituitary (10) and, possibly, may be caused by the increase in inhibin secretion. This substance, still hypothetical in humans, has been isolated in bovine follicular fluid (12). Since FSH causes the preferential growth of the granulosa cells, the hypothesis that these cells provide a feedback signal to control its secretion, appears logical. The once proven existence of inhibin would also provide an excellent explanation for the differential secretion of FSH and LH (13).

Experimental studies suggest that the pituitary gonadotropes possess the capacity to synthesize and release FSH, while the LH production appears dependent upon GnRH stimulation. The studies with synthetic GnRH have demonstrated the existence of two pools of gonadotropins. The first, representing the pituitary sensitivity, is acutely releasable. The second, pituitary reserve, requires sustained GnRH stimulation. The sum of these two pools is called pituitary gonadotropin capacity (13). Pituitary capacity undergoes physiologic changes. During the early follicular phase, it is at the minimum. Towards the midfollicular phase the reserve pool increases. Henceforth to midcycle surge, pituitary sensitivity exceeds reserve. During the luteal phase, pituitary capacity declines.

It has been well established that the initiation of midcycle surge of LH depends upon the critical level of estradiol in the hypothalamic-pituitary circuit. Estrogen seems to be the inducer, but the LH-surge trigger may involve several complex biochemical steps, involving prostaglandins and GnRh-syn-

thesizing neurones. Laboratory evidence suggests that in the hypothalamus, dopamine and norepinephrine containing neuronal pathways affect peptidergic neurones containing the hypothalamic releasing and inhibiting factors (14).

The fascinating link between the catecholamines and GnRH activity is the presence of catecholestrogens in the CNS. Particularly, the hypothalamus is capable of *in situ* formation of 2-hydroxyestradiol and 2-hydroxyestrone (15, 16). Two-hydroxylation gives a catechol structure to the aromatic A-ring of the estrogen molecule, providing the resulting compound with two faces: a catechol side and the estrogen side. Such compound has the potential of interacting with both the catecholamines and the estrogens. Catecholestrogens may have a more rapid and direct extracellular mechanism of action, than the classical action of a simple steroid hormone (13).

It appears that the feedback regulation of GnRh release may be at least partially based upon CNS formation of catecholestrogens, that subsequently affect the biosynthesis of catecholamines. Furthermore, the feedback may be influenced by prostaglandins. The brain can synthesize and secrete these substances. Studies have demonstrated that Prostaglandin E causes the release of gonadotropins *in vivo*.

Although many aspects of the normal ovulatory cycle still remain unclear, we accept that for the normal function, dynamic synchrony within the CNS-hypothalamuspituitary-ovary axis must exist. Thus, when this synchrony is disturbed, the resulting pathophysiology is reflected most frequently by anovulation and menstrual aberrations. The integration of the brain with the peripheral endocrine system involves the long-loop feedback (steroids to the hypothalamus-pituitary), the short-loop feedback (pituitary-hypothalamus) and the suprahypothalamic connections to the centers of CNS controlling behavior, mood and the biologic rhythms affecting the maintenance of social and environmental adaptation and homeostasis (13).

It appears obvious that the prevalent cause of anovulatory states is the alteration in CNS-hypothalamic integration. Both animal and human observations imply the presence of a central mechanism to prevent reproduction in unfavorable circumstances. While these facts should be kept in mind during the evaluation process of anovulatory patients, one must entertain the possibility of organic causes which should be first ruled out.

While many classifications of anovulation have been suggested, for practical purpose we find the one outlined by Yen (20) to be most useful:

1. Hypothalamic chronic anovulation—resulting from altered CNS-hypothalamic integration and inappropriate GnRH secretion.
2. Pituitary chronic anovulation—caused by abnormalities of gonadotropes function.
3. Abnormal feedback—caused either by iatrogenic influences, excessive extragonadal estrogen production, abnormalities of SHBG, or androgen excess, may it be functional or due to a tumor.
4. Abnormalities of associated endocrine/metabolic systems—adrenal, thyroid, nutritional deficiencies.
5. Pituitary-hypothalamic dysfunction—psycho-neuroendocrine disturbances, functional or organic hyperprolactinemia, hypopituitarism, empty sella syndrome, isolated gonadotropin deficiencies.

In view of multiplicity of etiologic factors, the management of anovulation should never be empiric. Prior to any therapeutic effort, as precise a diagnosis as possible must be made.

In evaluation of anovulatory patients, besides a detailed history and examination that may be revealing, the total endocrine profile should include the levels of FSH, LH, prolactin and, in the presence of defeminization, free testosterone and DHEA-S. In addition, thyroid profile is usually obtained. Specific disorders are usually readily recognized, and their correction will result in restoration of ovulation. Based, however, on the endocrine profile, four patterns will most frequently evolve:

- I. Normogonadotropic, normoprolactinemic. This is probably the most commonly encountered pattern, based on static testing, and it indicates a nonorganic state of dysfunction.
- II. Normogonadotropic, normoprolactinemic with an altered LH/FSH ratio. This group represents patients with the polycystic ovaries syndrome. It is perplexing that, to this date, the precise pathogenesis of this syndrome remains unclear. It is interesting to speculate that it may originate as an adrenal disorder during the phase of sexual maturation (22). An exaggerated adrenarche, caused by stress or functional hyperprolactinemia may cause an adrenal androgen excess. In turn, peripheral conversion of these steroids increases the amount of estrogens that induce inappropriate gonadotropin secretion. Henceforth, nonphysiologic gonadotropin levels cause overproduction of ovarian androgens that close the vicious circle. Chronically ele-

vated LH observed in this syndrome may be caused by estrogens inhibiting FSH, preferentially.

- III. Hyperprolactinemia. With this finding, the presence of pituitary adenoma must be evaluated by the use of hypocycloidal polytomography.
- IV. Hypogonadotropic. The multiple causes of pituitary failure must be assessed by a complete endocrine workup, dynamic testing of pituitary function and by polytomography. Clinically, the most useful dynamic tests are:
 - a. GnRH test. Infusion of this hormone (100 mcg IV) produces normal response of LH peak in 30 and FSH in 45 minutes.
 - b. Response to Clomid. Following administration of 100 mg daily for five days, gonadotropins are determined on day 0 and 10, and plasma progesterone on day 21. For positive response, ovulation need not occur.
 - c. TRH test. Evaluation of TSH and PR response, following intravenous injection of 500 mcg, both hormones should peak in 30 minutes.
 - d. Chlorpromazine (CPZ) test. Assessment of PR secretion and reserve of the pituitary. Following IM dose of 25 mgm the rise in PR will occur in one to three hours.
 - e. Assessment of GH reserve. This includes insulin tolerance and arginine tolerance tests.
 - f. ACTH reserve metyrapone test.
- V. Hypergonadotropic group is indicative of ovarian failure.

Review of Therapeutic Methods in Anovulatory States

Patients in Groups I and II, who are desirous of childbearing, should be treated with individualized, graduated regimens of Clomid. The doses should be increased until the ovulatory response occurs. Ovulatory doses should be repeated until conception takes place. If, however, ovulatory response was present for three cycles and pregnancy did not occur, the reassessment of a male factor as well as the pelvic architecture of the patient is mandatory. Rare patients with polycystic ovaries syndrome, who fail to ovulate with Clomid, may be considered for the ovarian wedge resection. This is my personal therapeutic preference over the use of Pergonal in

these women, who may exhibit an extreme sensitivity to this drug. In performing the wedge resection, we have adopted the techniques from microsurgery, in order to minimize the tissue trauma, abrasions and subsequent adhesions formation. The hormonal effects of this operation have been well studied (23). The immediate effects include transient rise in progesterone, fall in estrogen and LH. Among later postoperative effects, the most important appears to be the reversal of LH/FSH ratio to normal, initiation of cyclic gonadotropins activity and restoration of ovulatory process.

Our personal experience with Clomid compares well with the general observation of ovulatory response in 70-80% of patients and the pregnancy rate of 30-40%. It is important to remember that although the majority of conceptions occur within the first three ovulatory cycles, significant number of patients will require longer duration of therapy, and there are no contraindications to continue the treatment in patients who respond with ovulation and who do not have any additional factors that may affect their ability to conceive.

In a recent report on gestational outcome of Clomid-related conceptions (24), a twinning rate of 12.8% appeared to be the most important complication. Spontaneous abortion rate was 26.5%. The birth defects incidence of 3.1% was not increased as compared with the population at large.

Patients in Group III, who are free of pituitary pathology, should exhibit an 80% ovulatory response when treated with ergocryptine. Since this agent exerts a direct effect on the hypothalamic-pituitary unit, it effectively decreases the synthesis and release of prolactin, allowing for restoration of ovulatory process. Patients with prolactin-producing pituitary adenomas must be evaluated carefully. Especially with the microadenomas, there is no unanimity of opinions as to the therapeutic modalities, but recent reports (25) indicate that ergocryptine may be used with relative safety.

Patients in Group IV, who exhibit the failure of gonadotropins, are candidates for ovulation induction with HMG (Pergonal). It is a good policy to establish, beyond doubt that their male partners have a normal sperm count, and that the patients have perfectly normal pelvic organs, and ovaries capable of response and documented tubal patency before the initiation of Pergonal therapy. With this agent, the key to a successful therapeutic outcome and avoidance of complications is an extremely close and careful clinical and laboratory monitoring of the response, a flexible dose of the medication tailored to the patient's response, and a triggering

of the ovulation with HCG at the optimal time—when the serum estradiol level is at least 50% superphysiologic, in a range of 600–1000 pg/ml.

In my series of 30 patients treated with Pergonal, ovulation rate was 99%. Twenty-seven of these patients conceived (90%). Of these, 20 had singleton pregnancies, two had twins, one conceived triplets (at this time 32 weeks of gestation), three had quadruplets (two sets living), and one aborted septuplets. Multiple courses of Pergonal were necessary in only eleven patients (33%).

In summary, we have attempted to outline the advances in theory and practice of diagnosis and management of anovulatory disorders. One point should come across: in dealing with the derangement of ovulatory cycle, one must be very basic and very honest. There is no way to mask our current ignorance, and we must make the best with what we know so far. Let us hope that the research of the future will bring us more information and insight into this exceedingly complex, exciting and frustrating process: ovulation in the human female.

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Orthostatic Hypotension in the Holmes-Adie Syndrome

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Abstract

A patient with orthostatic hypotension (OH) was found to have tonic pupils and areflexia. On investigation, findings indicative of the Holmes-Adie syndrome and an afferent baroreceptor block were found. There was no evidence of peripheral sensory or autonomic neuropathy, which can be confused with the Holmes-Adie syndrome. Afferent baroreceptor block should be considered part of the spectrum of nervous system involvement in the Holmes-Adie syndrome.

Holmes and Adie described tonic pupils with the absence of one or more deep tendon reflexes occurring *sui generis*, in the absence of neurosyphilis or peripheral neuropathy (1, 2). A variety of evidence has been accumulated that identifies the anatomic site for tonic pupils in the Holmes-Adie syndrome as the ciliary ganglion. Schie demonstrated constriction of tonic pupils with ocular dilute methacholine, which is taken as evidence for denervation supersensitivity (3). Ruttner (4) and Harriman and Garland (5) found degeneration of ciliary ganglion neurons at autopsy in patients with the Holmes-Adie syndrome. The anatomic site for the loss of phasic stretch reflexes is unsettled. Several electrical studies have noted absence or depression of the H-wave response with normal motor and sensory nerve conduction velocities. (6, 7). The possible sites for the selective abnormality of the H reflex include the presynaptic terminals of group 1a dorsal root fibers or internuncial neurons in the spinal cord (8). There is no clinical, electrical or anatomic evidence for peripheral nerve abnormality in the

Holmes-Adie syndrome. Orthostatic hypotension (OH), in association with disorders labelled as Holmes-Adie syndrome, has been noted previously. However, the triad of tonic pupils, areflexia and OH can occur in any peripheral neuropathy in which sensory and autonomic fibers are involved (9-12). This does not constitute the syndrome described by Holmes and Adie. It is essential that a thorough investigation is undertaken to rule out peripheral sensory and autonomic neuropathy before concluding that the Holmes-Adie syndrome is present. We wish to present our findings in a patient with tonic pupils, areflexia and OH.

Case Report

A 63-year-old woman was in good health until September 1975, when she developed a low grade fever associated with nausea and vomiting that lasted for three days. She then had several episodes of loss of consciousness while standing and was admitted to a hospital. The family history was negative and the patient did not consume alcohol. There was no history of diabetes mellitus or venereal disease and no history of difficulty with sweating, tear production, bowel or bladder function. Physical examination was normal except for orthostatic hypotension, with a drop in blood pressure from 150/80 mm Hg, sitting, to 90/60 mm Hg, standing. Skull x-ray electroencephalogram and brain scan were normal. An upper gastrointestinal series, barium

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enema and intravenous pyelogram were normal, as were chest x-ray and electrocardiogram (ECG). Routine laboratory studies were normal except for a hematocrit of 35%. Serum folate level was normal, serum iron level was low and serum iron binding capacity normal. The patient was discharged on iron supplements. The patient's OH did not improve and she was admitted to another hospital in January 1976. Her hematocrit was 30% and her hemoglobin 10.0 gm/100 ml, with normal red blood cell indices. Bone marrow was normocellular with increased iron stores. A stool guaiac was negative, serum haptoglobin was normal, and a Coombs test was negative. Liver, spleen and bone scans were normal. Twenty-four hour urine collection for ketosteroids was normal, as were thyroid function tests. A urine screen for Bence-Jones protein was negative. The patient was discharged on fludrocortisone acetate, 0.2 mg/day, and continued to have syncopal episodes. In February 1977, she had unequal pupils that were unreactive to light. On admission to The Mount Sinai Hospital in May 1977, her blood pressure (BP) was 120/80 mm Hg supine, and 60/20 mm Hg, standing. General physical examination was otherwise normal. Neurologic examination disclosed unequal pupils, 4 mm OD and 3 mm OS. Both were minimally reactive to light and constricted promptly to accommodation. Visual fields and fundoscopic examination were normal. The patient was totally areflexic even with reinforcement. No pathologic reflexes were present. Muscle strength, tone and mass were normal. Sensory examination to all modalities was normal. Routine blood chemistries, chest x-ray, ECG and urinalysis were all normal. The hematocrit was 31.8%, and hemoglobin 10.8 gm/100 ml with normal red blood cell indices. Reticulocyte count was 0.1% and the erythrocyte sedimentation rate was 38 mm/hour. Serum folate, B₁₂, iron and iron binding capacity were all normal. A Coombs test was negative. Stool guaiac was negative. Serum and cerebrospinal fluid VDRL were negative, as were cerebrospinal fluid cell count, protein and glucose. A 24-hour urine collection for 17-ketosteroids was normal, as was a 5-hour glucose tolerance test. Computerized axial tomography of the brain and electroencephalogram were normal.

Electromyographic data: Median and peroneal nerve motor conduction velocities, distal latencies and amplitudes of evoked potentials were normal. No H reflex was elicited by stimulation of the posterior tibial nerve at stimulation rates of one per second and one per two seconds, as well as following several short bursts of tetanic stimulation. On

needle electromyography, motor units were mildly decreased in duration and of normal amplitude. There was a mild increase in polyphasic potentials noted. The results were interpreted as possibly representing an early myopathic process. A gastrocnemius muscle biopsy was normal. Stains for amyloid were negative.

Autonomic Function Tests

Pupillary tests: Both pupils constricted to solutions of 0.125% and 0.0625% pilocarpine. Both pupils dilated normally to solutions of 10% cocaine and 1% hydroxyamphetamine. On slit lamp examination there was bilateral segmental palsy of the iris sphincter to light. The results indicate cholinergic denervation of the pupil at or distal to the ciliary ganglion with intact sympathetic innervation.

Tilt table tests: The BP decreased from 120/80 mm Hg supine, to 60/20 mm Hg on 90 degree tilt. The pulse remained constant at 72/min on full tilt. In response to 60 seconds of an ice water stimulus, the supine BP increased from 120/80 mm Hg to 150/100 mm Hg, and the pulse increased from 72/min to 84/min. Digital blood flow, measured with a finger plethysmograph coupler on a Beckmann R511 dynagraph recorder, showed normal vasoconstriction during the pressor test. The results indicate that the efferent sympathetic pathway is intact.

Catecholamine histofluorescence assay: A muscle specimen was stained for catecholamine histofluorescence of perivascular nerve endings using the Falck-Hillarp technique (13). There was normal catecholamine histofluorescence on all sections with this technique (Fig. 1).

Norepinephrine (NE) infusion: Dilute NE, 5–45 ng/kg/min, was infused intravenously with a Harvard pump. BP and pulse were monitored continuously. The infusion rates are below that required to raise BP in normal adults. There was a 25% increase in systolic BP at the higher infusion rate. There was no reflex bradycardia induced by the increase in BP. The degree of supersensitivity is much less than that observed in postganglionic denervation (14).

Resting Plasma NE level: Plasma NE was drawn from an indwelling catheter after the patient was recumbent for two hours. The level was 365 ng/ml, within the normal range for this age group (15).

Valsalva Maneuver: The patient performed the valsalva maneuver by maintaining an expiratory pressure of 40 mm Hg for ten seconds while supine. Heart rate was recorded on an ECG for 20 seconds



FIG. 1: Catecholamine histofluorescence. Cross section of a blood vessel passing between muscle fibers. The arrows indicate brightly fluorescing adrenergic nerve terminals.

before, and 30 seconds after expiration. The normal overshoot in BP, following forced expiration, results in a reflex-induced bradycardia that was quantitated using previously published methods (16). There was no bradycardia induced with valsalva maneuver.

Oculocardia reflex: Pressure to the eyeballs produces a bradycardia mediated by the vagus nerve that is independent of afferent baroreceptor function (17). Application of pressure to both eyeballs for 30 seconds resulted in a prompt decrease in pulse of 11%.

Sweat test: Whole body heating with a heating blanket for thirty minutes resulted in normal sweat production.

Discussion

The autonomic function studies identified the afferent baroreceptor as the site of the lesion responsible for the OH. The normal cold pressor test and normal catecholamine histofluorescence of ad-

renergic plexuses on muscle biopsy indicate a normal efferent sympathetic pathway. Patients with OH caused by peripheral autonomic neuropathy have depleted peripheral catecholamine stores (18–20). The normal plasma NE level at rest is consistent with an intact peripheral adrenergic system, as patients with peripheral adrenergic denervation have been reported to have low resting plasma NE levels (21). The baroreceptor reflex was absent on standing, in the valsalva maneuver and during the NE infusion, in which no reflex bradycardia occurred. The lesion was isolated to the afferent arc of the baroreceptor by the demonstration of vagus-induced bradycardia in the oculocardiac reflex, which indicated that the efferent arc of the baroreceptor pathway was functioning. There was no evidence for involvement of peripheral motor, sensory or autonomic nerves in this patient, though the possibility of an early steroid myopathy was raised by the electrical studies. The supersensitive response to dilute cholinergic eyedrops and the selectively absent H reflex fulfill the criterion for diagnosis of the Holmes-Adie syndrome.

TABLE I

The Syndrome of Tonic Pupils, Areflexia and Orthostatic Hypotension: Site of Lesions

Problem	Holmes-Adie Syndrome	Peripheral Sensory and Autonomic Neuropathy
Tonic pupils	Ciliary ganglion	Postganglionic cholinergic fibers to pupil
Areflexia	?Group 1a dorsal root fibers * ?Spinal internuncial neurons	Peripheral sensory nerves
Orthostatic Hypotension	Afferent baroreceptor	Postganglionic adrenergic fibers to perivascular plexuses

Several reports of abnormal circulatory reflexes in the Holmes-Adie syndrome have appeared. In most cases, adequate studies to rule out peripheral neuropathy or neurosyphilis were not done. In some previous reports of OH in association with the Holmes-Adie syndrome, electromyographic studies to rule out peripheral neuropathy were not performed (22, 23). Berkowitz and Zweifach, in 1970, reported abnormal responses to the valsalva maneuver in 11 patients with tonic pupils (24). Four of the patients had positive serologic tests for syphilis or a previous history of syphilis. In three patients, serologic tests for syphilis were not performed. Of the remaining four patients, one had an alcoholic neuropathy and another had diabetes. Similarly, reports of abnormal sural nerve biopsies (25) and of progressive peripheral neuropathy (26, 27) in the Holmes-Adie syndrome actually describe tonic pupils and areflexia as part of a diffuse neuropathy with a prominent autonomic component (Table I).

Only one previous report of OH in the Holmes-Adie syndrome has adequately documented the absence of peripheral neuropathy with appropriate electromyographic studies. Johnson et al in 1971 (28) reported two cases with OH caused by what was thought to be afferent baroreceptor block. Physiologic and pharmacologic studies pointed to a defect in the afferent arc of the baroreceptor reflex, similar to what was found in our patient. While baroreceptor dysfunction in diffuse autonomic neuropathies is common (29), isolated involvement of the afferent arc of the baroreceptor pathway is a unique cause of OH that has only been described in the Holmes-Adie syndrome. The spectrum of nervous system involvement in the Holmes-Adie syndrome should be extended to include afferent baroreceptor dysfunction.

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Neurologic—Neurosurgical Notes

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Parasagittal Subdural Empyema: A Case Report With Computerized Tomographic Scan Documentation

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Abstract

A 10-year-old Haitian male developed pan-frontal sinusitis that culminated in right focal seizures, progressive lethargy and mild persisting right hemiparesis. His cerebrospinal fluid profile was consistent with meningoencephalitis. Antibiotic therapy was initiated and exenteration of the left frontal, ethmoidal and maxillary sinuses performed. A computerized tomographic (CT) scan of the brain on admission was normal. One month later, a large left parafalcine lucency was found. A separate lucency was apparent in the region of the left temporooccipital junction. Both lucent areas were surrounded by thickened enhancing capsules. No clear documentation of communication between these cavities could be discerned on the horizontal cuts. Drainage of a large subdural empyema was accomplished by left parietooccipital craniotomy. The duration of postoperative antibiotic therapy with Cephalothin and Carbenicillin was 22 days. Cephalexin was continued orally after discharge for an additional two weeks. Cultures of the maxillary sinus pus grew *staphylococcus albus*. Microaerophilic streptococcus was seen on cultures of the subdural pus. The patient made a complete recovery. The follow-up CT scans showed complete resolution of the subdural cavity and its enhancing capsule confirming the suspicion that both cavities were in continuity.

Introduction

Despite newer diagnostic techniques and antibiotics, the mortality rate from subdural empyema still ranges from 15% to 43% (11). Factors contributing to these figures include failure of prompt diagnosis, delayed or inadequate surgical intervention, the onset of cortical vein thrombophlebitis, subadjacent cerebritis, the concomitant existence of a brain abscess and dural sinus thrombosis. Lastly, failure to debride the source of infection may lead to recurrence and/or extension of the infection.

The compelling triad of sinusitis, seizures and focal neurologic deficit should lead to persistent

investigation to discover a subdural empyema. The CT scan is of paramount importance in facilitating this diagnosis.

The case presented here is a unique example of the value of repeat CT scanning in following the development of subdural empyema and its resolution after antibiotics and surgery. In addition, the unusual instance of an exclusively unilateral parafalcine subdural empyema extending onto the ipsilateral tentorial leaflet is described.

Case Report

The patient, a 10-year-old right-handed Haitian male was admitted with a two-day history of pain in the left supraorbital region, vomiting, fever of 102° F., progressive obtundation and right-sided seizures. General physical examination disclosed swelling and tenderness of the left periorbital tissues and a Grade 2/6 systolic ejection murmur.

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There was resistance to flexion of the neck, but no definite Brudzinski sign was demonstrated. Neurologically, he was stuporous, responding to deep pain purposefully and vigorously in all extremities. The peripheral WBC was 7,200, ESR 25, sickle cell prep. negative. Cerebrospinal fluid showed 340 WBCs with 96% polys, 4% lymphs and 10 RBCs. The protein was 90 mgms % and the glucose was 86 mgms % with blood sugar of 185 mgms%. Skull films showed opacification of the maxillary, ethmoidal and frontal sinuses, the left greater than the right. An EEG showed diffuse cerebral dysfunction bilaterally. The initial CT scan was read as being within normal limits (Fig. 1). Chloramphenicol 90 mgms every six hours and aqueous penicillin 1.5 million units every six hours were begun intravenously.

The day after admission, repeat cerebrospinal fluid examination showed 150 RBCs and 260 WBCs. The protein was 156 mgms % and glucose 60 mgms %. Blood and CSF cultures were negative. He began to respond to verbal stimulation. Further improvement was noted over the ensuing three days, but the swelling of the left periorbital tissues progressed and a mild right hemiparesis along with a mild left sixth nerve paresis became apparent. Because of the persisting fever and periorbital swelling, a left Caldwell-Luc procedure was performed with radical antrostomy and ethmoidectomy. Cultures of the sinus pus grew staphylococcus albus. The right hemiparesis became more evident with lower extremity weakness exceeding that of the upper extremity. Ten days after admission, a technetium 99 brain scan showed no abnormal uptake. A repeat EEG demonstrated left hemispheric dysfunction to a much greater extent than right, indicating a deterioration from the initial tracing. A repeat cerebrospinal fluid examination 15 days after admission

showed 15 WBCs and 2 RBCs. The protein was 21 mgms % and glucose 66 mgms %.

In view of the persisting fever, the antibiotics were changed on the sixteenth postadmission day to Cephalothin and Carbenicillin. He became afebrile. In the following two weeks, he complained of diplopia and experienced several episodes of vomiting. On the twenty-seventh day, fundoscopic examination disclosed papilledema. The CT scan was repeated with contrast and showed a holohemispheric curvilinear left parasagittal lucent cavity with an additional cavity in the region of the left occipitotemporal junction (Figs. 2-6). Definite communication between the cavities could not be ascertained on the horizontal cuts. Enhancing capsules surrounded both cavities. The parasagittal cavity appeared to extend up beyond the parasagittal angle to the hemispheric dura (Figs. 6a, b). Surgical drainage was undertaken to rule out a parasagittal empyema.

A left parietooccipital craniotomy was performed. After lateral mobilization of the occipital and parietal lobes from the falx, the parasagittal subdural cavity was encountered—a distance of four centimeters from the internal occipital protuberance. The cavity was densely adherent to both the brain and the falx and multiple satellite nodules of granulation tissue were scattered along the falx. Aspiration of 60 cc of pus through a large bore catheter was followed by copious irrigation with a bacitracin solution. Attempts to reach the tentorial cavity were thwarted by adhesions between the cortex and the tentorium. A second attempt to enter the parasagittal cavity through a left frontal burr hole and craniectomy disclosed normal cortex capping the cavity at the frontal parasagittal angle. Satisfactory decompression of the hemisphere was achieved. Postoperatively, the catheter was withdrawn over a three-day period. Intravenous Carbenicillin and Cephalothin were continued for 22 days and Cephalexin was given orally after discharge for an additional two weeks. Cultures of the pus yielded a light growth of microaerophilic streptococcus sensitive to the antibiotics used.

The patient made an excellent recovery with resolution of all neurologic dysfunction. He became fully ambulatory and could return to school. His EEG reverted to normal. Repeat CT scans at 20 days and three and one-half months postoperatively showed disappearance of both cavities, confirming their continuity and disappearance of the enhancing capsule except for a small area on the left tentorial leaflet (Figs. 7-9). Mild ventricular enlargement was seen and was followed with successive scans.

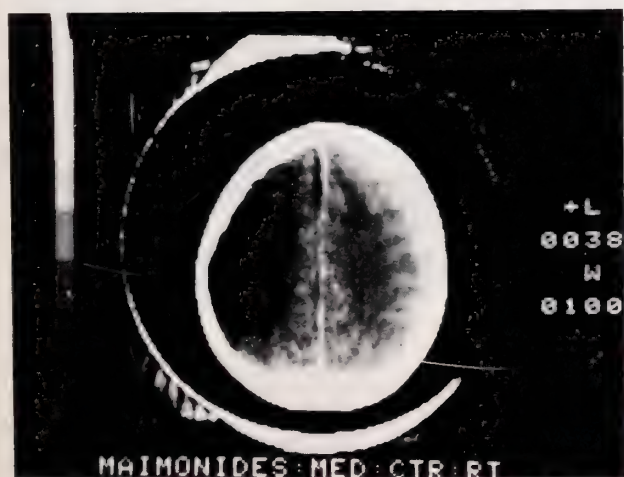


FIG 1. Scan with contrast on admission.

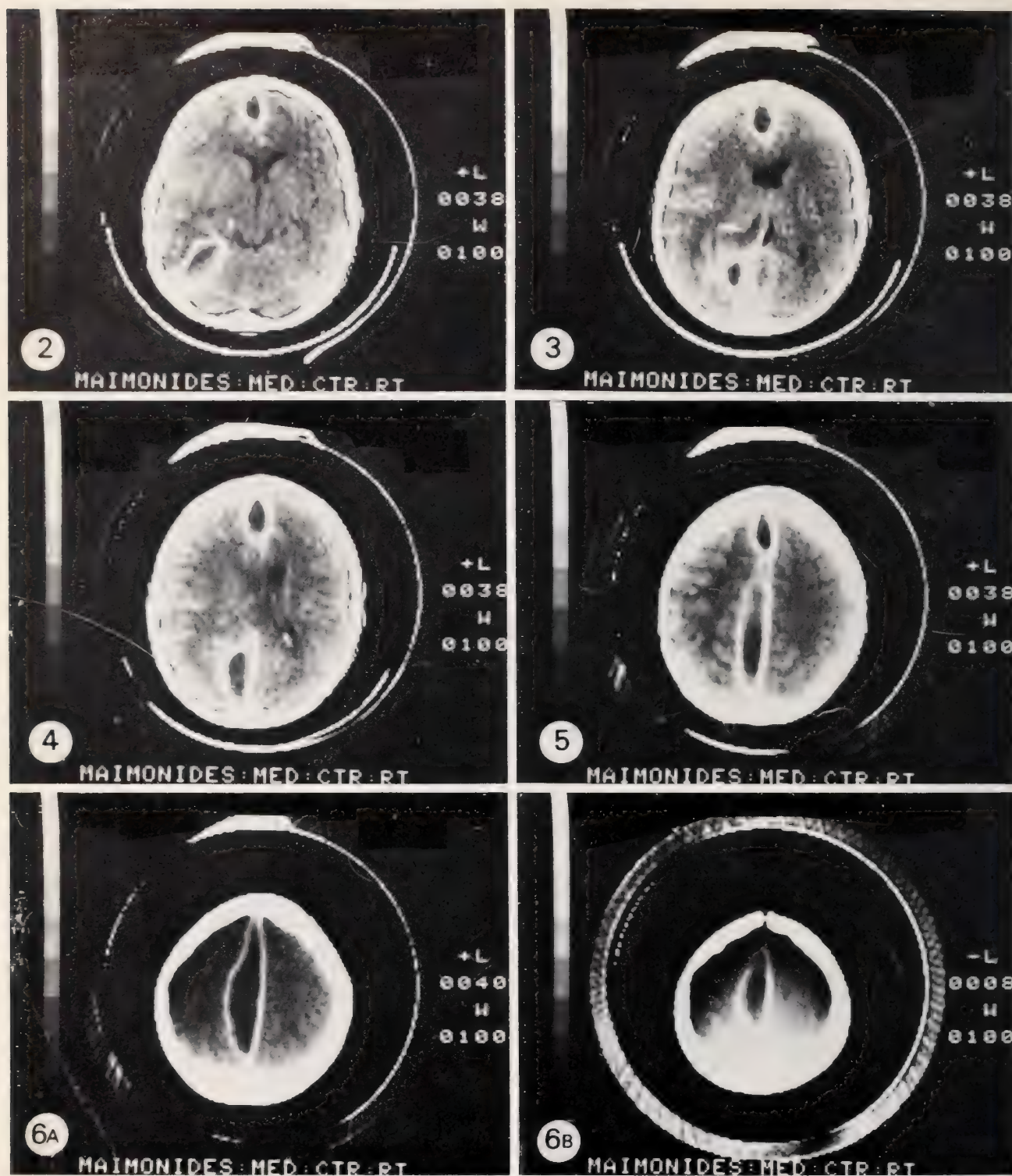
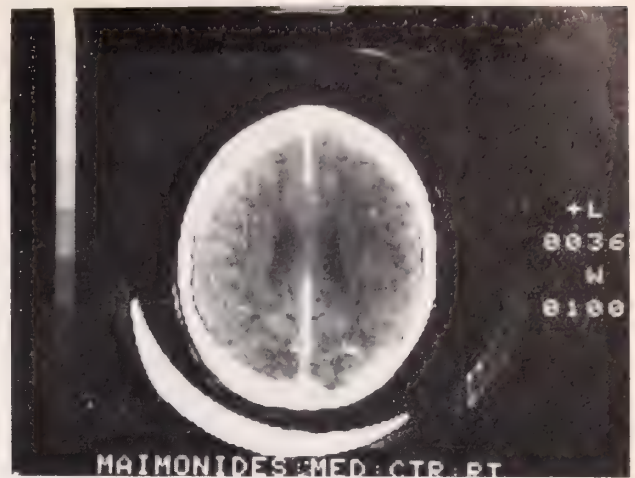
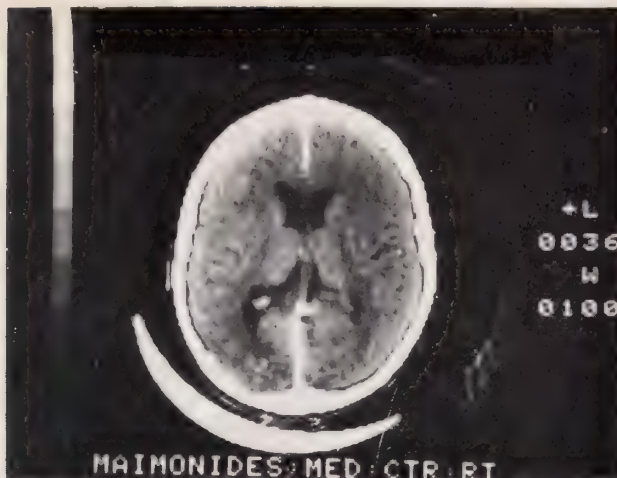


FIG 2-6. Subdural cavity preoperatively.

Discussion

The morbid anatomy (7, 24), pathogenesis (4, 7, 10, 13, 19, 23, 24), angiographic appearance (11, 12,

15, 21, 22, 27), antibiotic therapy (6, 13), surgical therapy (4, 13, 16-18, 26), and complications (6, 7, 13, 17-19, 24) of subdural empyema has been thoroughly discussed. Reports of CT scan documenta-



FIGS. 7 and 8. Twenty days postoperatively without contrast.

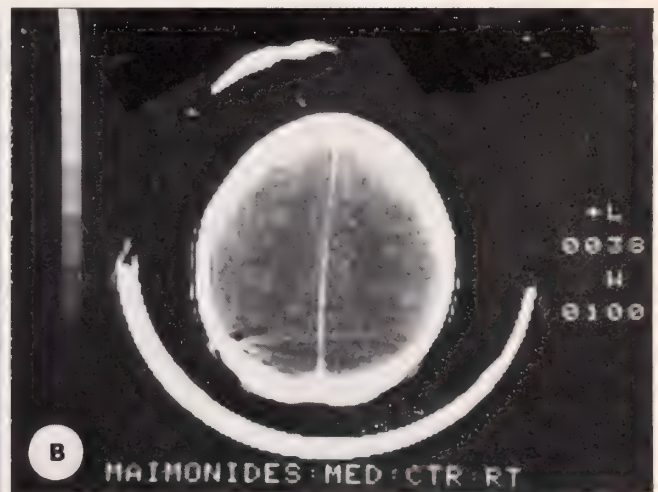
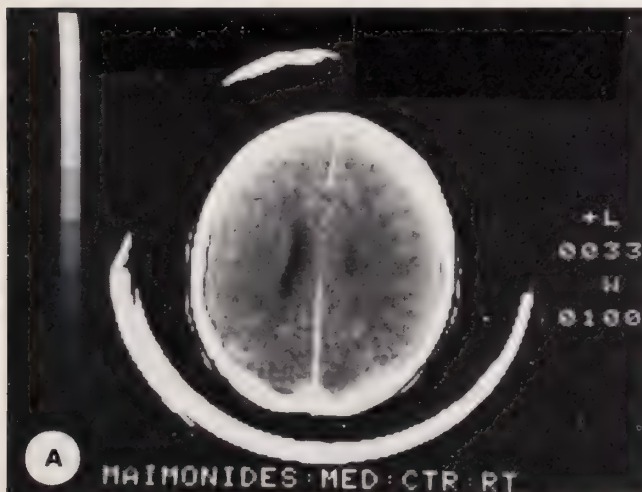


FIG 9. Three and one-half months postoperatively with contrast.

tion of the evolution and regression of subdural empyema during the course of antibiotic and surgical management are scarce (5, 22).

Exclusively, parasagittal primary location of subdural pus is uncommon. Although reports cite incidences of subdural empyema of 13% to 21% of all localized intracranial bacterial infections (6, 13), reports of primary parasagittal empyemas are rare. Wilkins and Goree (27) reported 2 of 12 cases over an 11-year-period. List cited one instance (18). Keith (14) mentioned 3 of 6 instances, but in only one case was the empyema primarily in the parasagittal location. Courville (7) found 4 of 42 and Kaufman et al (13) found 3 of 17 cases. The cases of the latter two authors represent secondary extension into the parasagittal area from a hemispheric subdural collection. This sequence is by far the more common (18).

Otorhinologic infections significantly exceed

other sources, including operative intracranial infections and meningitis (13). Hematogenous spread and infection of sterile subdural effusions are singularly rare events (10, 13, 28). Whether osteomyelitis and external pachymeningitis necessarily precede the formation of a subdural abscess or whether organisms are conveyed centripetally via progressive thrombophlebitis of mucosal, emissary, cortical veins and the intradural venous plexus is not established with certainty. The precise manner by which bacterial organisms gain access to the potential subdural space is not known (28).

There does not seem to be a single factor that helps in understanding why in some cases, the primary subdural empyema is exclusively parasagittal and, in other cases, primarily hemispheric. Proximity plays a certain rôle. The circular or petrosal sinuses may be involved following sphenoid sinusitis leading to a perihypophyseal abscess (3, 25)

and pus may extend onto the tentorium following mastoiditis (19). Botterell and Drake (4) concluded that infection of the ethmoidal and frontal sinuses could spread centripetally in any one of three directions: deep under the orbital surface of the frontal lobe and along the Sylvian fissure, medially along the falx ultimately to overlie the tentorium, and superolaterally over the convexity. Once pus gains entry to the subdural space, prior to the adherence between the inner mesothelial surface of the dura and arachnoid, it may spread from one end of the neuraxis to the other. Ray and Parsons (23) described subfalcial spread. Schiller et al demonstrated pus in the spinal subdural space (24).

From our case and the one described by List (18) a pattern emerges that is typical for the primary parasagittal subdural empyema. In the acute phase with extensive subadjacent cerebritis and cortical vein thrombophlebitis or sinus thrombosis, the clinical course may be brief and irreversible. With the use of antibiotics, a subacute and chronic course ensues. The parafalcine collection enlarges and extends onto the ipsilateral tentorium under the occipital and posterior temporal lobes. Clinically the "eloquent areas" of cortex affected are the paracentral lobule, the cuneus and the lingual gyrus. Involvement of the paracentral lobule leads to crural greater than brachial hemiparesis and to seizures involving the lower extremity preferentially. Involvement of the cuneus and lingual gyrus may lead to homonymous hemianopia (18).

Whereas burr hole drainage may suffice for evacuation of subdural pus overlying the convexities, we concur with LeBeau (16, 17) that craniotomy should be the procedure of choice with parasagittal collections. Systemic antibiotics are mandatory. List (18) and others who performed burr hole aspirations often were required to pass a brain needle through normal cortex in order to enter a parafalcine cavity. Pus was then evacuated through normal brain. This technique carries the potential hazards of inadequate drainage, cortical damage, and damage to bridging cortical veins. As LeBeau has pointed out the parasagittal empyema cavity is thickest in the frontal region; therefore, it is less amenable to penetration by a probing blunt needle. An occipital approach is more appropriate from the standpoint of draining both parasagittal and tentorial cavities, should that be required. In our case, the depth of the cavity from an occipital approach was surprising, based upon the appearance of the CT scan (Figs. 2, 5). The depth of the cavity from the internal occipital protuberance was four centimeters. The granulation tissue along the falx and the ad-

herence of the medial aspect of the hemisphere to the capsule would have made burr hole drainage extremely difficult. Despite the CT scan appearance that the cavity extended up to and above the parasagittal angle (Fig. 6), it was clearly capped by normal cortex.

Our case raises an interesting point regarding antibiotic therapy. Namely, to what extent does the subdural capsule obtain its blood supply strictly from dural vessels or across the blood-brain or blood-CSF barrier on the arachnoidal side? Handa et al (11) recently noted the enlargement of the tentorial artery sometimes referred to as the artery of Bernasconi and Cassinari (1) in a case of a tentorial subdural empyema. Similarly, Kim et al (15) recognized enlargement of the anterior falx artery in subdural empyema. This suggests dural and leptomeningeal hypervascularity that should permit passage of antibiotics not known to cross the blood-CSF or blood-brain barrier into subdural empyema cavities and their capsules. An antibiotic such as Cephalothin may be efficacious in the treatment of subdural empyema but may not have significant value in the management of a brain abscess.

Because of the likely existence of microaerophilic organisms and because of high CSF and brain penetration we used chloramphenicol and penicillin initially. When the response was incomplete, we switched to cephalothin and carbenicillin. Preoperatively, the patient became afebrile. Postoperatively, there was virtually complete resolution of the empyema capsule (Figs. 7-9), justifying the use of this combination.

The inestimable value of the CT scan in interpreting the evolution and regression of subdural cavities is obvious. Coronal scanning was not available to us, but clearly would have helped in establishing preoperatively that the apparent separate cavities and capsules were, in fact, in continuity.

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Ophthalmologic Notes

Keith M. Zinn, M.D., Editor

Subretinal Neovascularization and Ocular Candidiasis

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Introduction

Fibrovascular proliferation from the choroid is a nonspecific, yet frequent manifestation of macular disease. These vessels are commonly referred to as subretinal neovascularization (SRN) and are associated with an increasing number of macular disorders, including hereditary diseases, congenital defects, degenerations, tumors and inflammations.

There have been a number of reports in the literature concerning the relationship between systemic candidemia and candida endophthalmitis (2-18). However, ocular candidiasis has not yet been reported to be associated with subretinal neovascularization. The following is a case report that describes subretinal neovascularization and ocular candidiasis originating from a primary chorioretinal inflammatory lesion in a patient with systemic candidiasis.

Case Report

E.Z. is a 63-year-old Caucasian woman with hyperthyroidism of one-year duration who was hospitalized in August 1976 for a large pelvic mass. Medical workup revealed a right renal cyst and a dermoid cyst of the ovary on the right side evidenced by an intravenous pyelogram, a nephrotomogram and gynecologic examinations. The patient developed systemic candidiasis approximately two weeks after admission that was documented by three separate blood cultures and one urine culture. An indwelling blood catheter tip also was positive

for candida albicans. Several days later the patient developed blurred vision in the right eye and the eye findings at that time were as follows:

Visual acuity with correction was 20/20⁻² in the right eye and 20/20⁻³ in the left eye. The extraocular motility studies revealed a full range of motion of either globe in all standard fields of gaze and the patient was orthophoric for distant as well as for near targets. The lids, ocular adnexae, and lacrimal gland apparatus were within normal limits in both eyes. Cranial nerve function II, III, IV, V (ophthalmic division) VI and VII were grossly intact bilaterally. Formal visual fields were not performed at that time, but confrontation fields were normal in both eyes. The conjunctivae, corneae and sclerae were within normal limits in both eyes and there was 1+ corneal arcus senilis in both eyes. Slit lamp examination of the anterior chambers revealed them to be clear and of moderate depth. There were Grade III open anterior chamber angles in both eyes. There was 1+ pigmentation of the trabecular meshwork in both eyes. The lenses were essentially clear with (1+) nuclear sclerosis noted O.U. The right vitreous cavity and 1+ cells and was hazy with a decreased foveal reflex. The left vitreous cavity was clear and unremarkable. Funduscopic examination revealed well demarcated optic nervehead borders in both eyes. The cup/disc ratio, in the horizontal meridian, was 0.2 O.U. The vein artery ratio was 3.2 O.U. Three focal zones of white exudate were observed in the neurosensory retina. Two of the lesions were temporal and one lesion was nasal to the optic nervehead in the right eye. Each lesion was about one-quarter to one-third of a disc diameter in size. The retinal lesions were edematous and were interpreted as patches of chorioretinitis in the right eye. No such lesions were seen in the left eye. Over the next 96 hours, the vitreous in the right eye became more cloudy, evolving into a panuveitis. At this point it was believed

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that the patient had embolic chorioretinitis secondary to systemic candidemia. The patient had been started on systemic Amphoterecin B and Prednisone, with topical Gentamycin and Chloromycetin medications. The regimen was maintained for several weeks. During this interval, the visual acuity decreased to the 20/200 level in the right eye. After

six weeks of therapy, the vitreous cavity cleared and the chorioretinal lesions looked quiescent and the Amphoterecin B was discontinued. Several days later the retinal lesions became active once again and the patient was restarted on systemic Amphoterecin B for an additional ten weeks. During this latter period, the retinal lesions grew in size, affect-

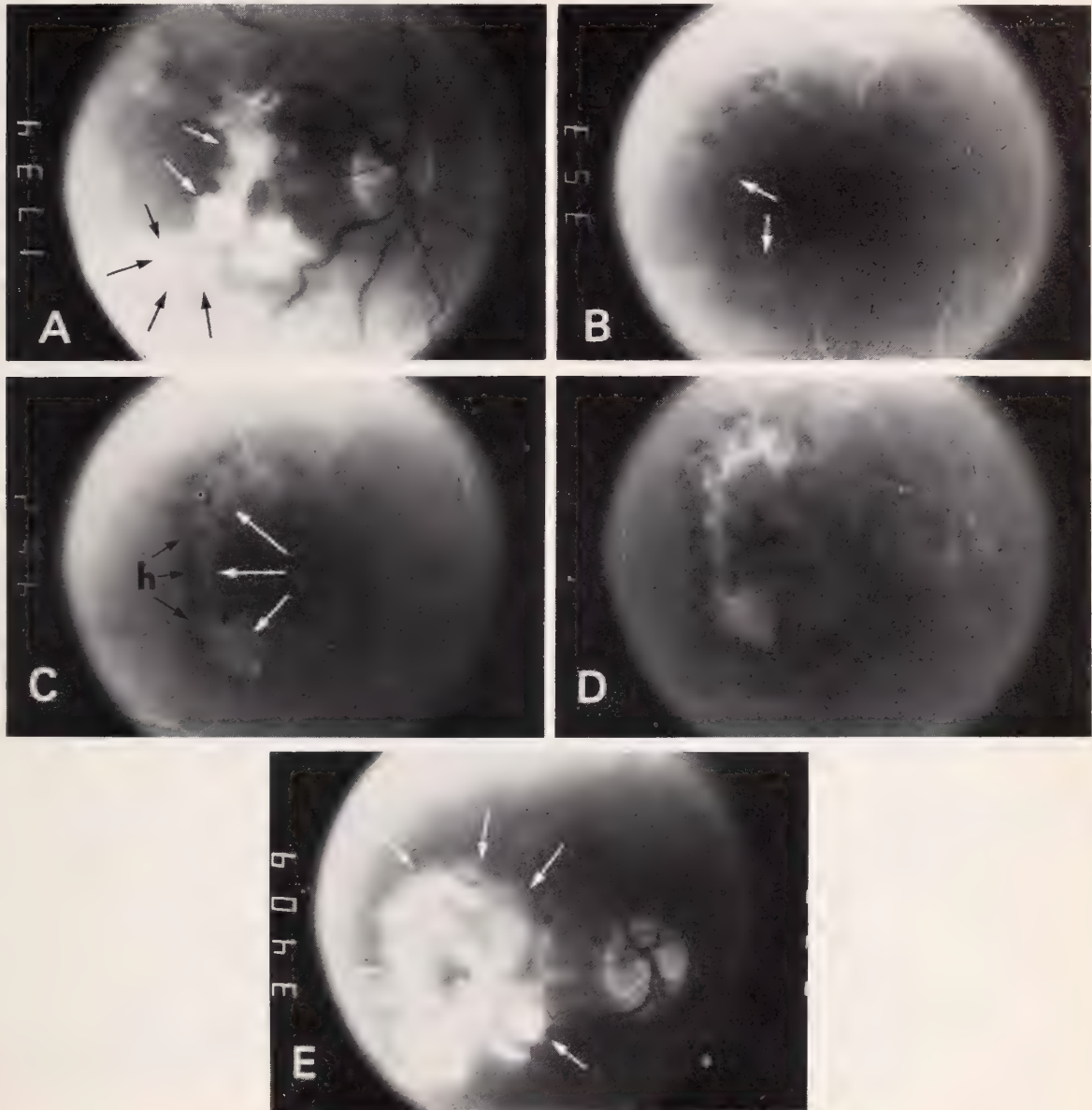


FIG. 1-A Fundus photo of the right eye. Note the preretinal glial tissue in the superior nasal macula (white arrows) associated with a vitreoretinal interface disturbance. Subretinal fibrosis is present in the nasal macula, coursing through the papillo-macular bundle. A greyish subretinal membrane is present in the temporal macula (black arrows) associated with an overlying exudative detachment of the sensory retina and subretinal hemorrhage.

FIG. 1-B Early arteriovenous phase of fluorescein angiogram with subretinal vessels beginning to fill with fluorescein (arrows).

FIG. 1-C Early venous phase angiogram. Note the large net of subretinal vessels at the temporal, superior and inferior aspects of the scar (white arrows). A zone of hypofluorescence is evident at the terminal aspect of the subretinal neovascularization corresponding to subretinal hemorrhage (H) and exudate.

FIG. 1-D Late arteriovenous phase. Note increasing fluorescence of subretinal neovascular membrane.

FIG. 1-E Late phase fluorescein angiogram. Note the late hyperfluorescence or leakage subretinally from the neovascular membrane into the subretinal space and into the scar tissue (white arrows).

ing the macular region in the right eye with a resultant central scotoma. The chorioretinitis decreased and the Amphoterecin B was finally discontinued. The vision had decreased to the counting finger level and a posterior subcapsular cataract was noted for the first time in the right eye.

Several months later, the patient developed open angle glaucoma in the right eye that was controlled with oral Diamox and topical Timoptic 0.5% medications. In addition, the patient had an associated uveitis in the right eye, but the chorioretinal lesions did not appear to be active on ophthalmoscopic examination. The uveitis was controlled with topical Decadron 0.1% in the right eye. Systemic steroids were avoided for fear of reactivating the candidiasis.

Fluorescein angiography studies were performed on the right eye and revealed subretinal neovascularization within the macular region. (Fig. 1).

Discussion

Subretinal neovascularization usually starts with a break or discontinuity of Bruch's membrane which is followed by an ingrowth of capillaries from the choriocapillaris. The new capillaries grow under the basal layer of the retinal pigment epithelium. Frequently, these vessels compromise the overlying

TABLE I

Ophthalmoscopic Findings Suggestive of the Presence of Subretinal Neovascularization

Dirty gray or yellow-green lesion
 Pigmented ring
 Hemorrhage (retinal, subretinal, sub-RPE)
 Exudate (retinal, subretinal, sub-RPE)
 Neurosensory retinal detachment $\pm$ turbid fluid
 Retinal pigment epithelium detachment
 Cystoid macular edema

TABLE II

Fluorescein Angiographic Findings Suggestive of the Presence of Subretinal Neovascularization

Vascular phase
 Early, lacy, nodular or irregular hyperfluorescence
 Extravascular phase
 Late hyperfluorescence (leak)
 Dense staining (fibrovasc. membr.)
 Fuzzy fluorescence (turbid fluid subret. space)
 Cystoid edema (overlying retina)
 RPE detachment
 Hot spot
 Meniscus hemorrhage
 Hypofluorescence
 Secondary to hemorrhage or pigm. ring surrounding Vessel complex

retinal pigment epithelium and invade the subretinal space (1). At this point a number of sequelae are possible, including hemorrhage, exudate formation, and development of fibrous tissue that may lead to a disciform scar. The latter reaction is usually associated with destruction of the outer layer of the neurosensory retina with concomitant loss of vision, especially within the macular region.

The clinical diagnosis of subretinal neovascularization is suggested by the presence of subretinal hemorrhage, exudate, intraretinal cystoid edema with neurosensory retinal detachment, or a dirty grey subretinal membrane (Table I). The subretinal neovascularization is confirmed by early lacy hyperfluorescence and late leakage on the fluorescein angiogram (Table II). The known macular disorders associated with subretinal neovascularization are listed in Table III. The list includes numerous gran-

TABLE III

Subretinal Neovascularization Differential Diagnostic Elements

Involucional Macular Degeneration
 Inflammation
 Presumed Ocular Histoplasmosis
 Toxocara
 Toxoplasmosis
 Cryptococcosis
 Sarcoidosis
 Candida
 Harada's Disease
 Serpiginous Choroidopathy (GHPC)
 Amppe
 Rubella
 Papilledema
 Long Standing Subretinal Exudate
 Trauma
 Choroidal Rupture
 Photocoagulation
 Idiopathic
 Tumors
 Choroidal Nevus
 Choroidal Malignant Melanoma
 Choroidal Osteoma
 Choroidal Hemangioma
 Choroidal Metastasis
 Retinal Pigment Epithelial Hypertrophy
 Retinal—Retinal Pigment Epithelial Hamartoma
 Hereditary
 Angioid Streaks
 Optic Nervehead Drusen
 Myopia
 Best's Disease
 Stargardt's Disease
 Polymorphic Dystrophy
 Retinitis Pigmentosa
 Dominant Drusen
 Sorsby's Maculopathy
 Dominant Subretinal Neovascularization
 Coloboma of the Optic Nerve

ulomatous ocular diseases, such as sarcoidosis, toxoplasmosis, toxocariasis and cryptococcosis.

The typical histopathologic lesion in candida chorioretinitis consists of a basic suppurative and granulomatous reaction containing epithelioid giant cells. The lesions tend to be focal, beginning in the choroid and breaking through Bruch's membrane, the retinal pigment epithelium and neurosensory retina and extending into the vitreous body (11, 14, 20). Fungi can be present at all zones of the focal lesion, and abscess formation can take place. Since a number of other granulomatous disorders affecting the eye have been associated with subretinal neovascularization, it is not surprising that a patient with ocular candidiasis, involving the macula, eventually develops subretinal neovascularization, resulting from the original inflammatory lesion. The embolic chorioretinitis in our case most probably resulted in a degenerative membrane-choriocapillaris complex. This, in turn, led to a subretinal fibrovascular ingrowth typical of all the macular diseases associated with disciform scarring as listed in Table I.

Summary

This is the first case report of ocular candidiasis associated with subretinal neovascularization in the macula. Following the initial chorioretinitis a subretinal fibrovascular ingrowth occurred leading to disciform scarring.

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UNUSUAL PROBLEMS IN SURGERY

A. ROBERT BECK, M.D., EDITOR

Total Intestinal Aganglionosis

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Abstract

Total intestinal aganglionosis is a rare condition, with only 11 instances reported in the literature. A case of aganglionosis extending to the distal stomach is reported and the literature is reviewed. It is of interest that there were three pairs of siblings in the 12 cases, giving rise to the concept that total intestinal aganglionosis is a distinct, inherited entity.

Hirschsprung's disease is usually limited to the rectosigmoid. Numerous instances of involvement of the entire colon and various portions of the ileum have reported (1-3). The first case of total intestinal aganglionosis was described by Bodian in 1951 (4).

involvement of the duodenum, and four with aganglionosis of the jejunum (Table I).

In the following case, there were no ganglion cells present in the myenteric plexus of the colon, small bowel or distal stomach.

TABLE I
Aganglionosis of the Entire Bowel Reported Cases

Author and Year	Sex	Affected Siblings	Extent of Disease
1. Bodian et al, 1951 (4)	M	Sibling of case 5	Distal duodenum to rectum
2. Lee, 1955 (5)	?	?	Duodenum to rectum
3. Boggs and Kidd, 1958 (6)	M	Sibling of case 4	D-J flexure to rectum
4. Boggs and Kidd, 1958 (6)	F	Sibling of case 3	D-J flexure to rectum
5. Bodian and Carter, 1963 (7)	F	Sibling of case 1	Duodenum to rectum
6. Walker et al, 1966 (8)	M	none	3rd portion of duodenum to rectum
7. Ahmed et al, 1971 (9)	F	Sibling of case 11	Duodenum to rectum
8. Asch et al, 1972 (1)	F	none	Entire duodenum to rectum
9. Talwalker, 1976 (10)	M	1 sibling died or intestinal obstruction	D-J flexure to rectum
10. Talwalker, 1976 (10)	M	none	D-J flexure to rectum
11. MacKinnon and Cohen, 1977 (11)	M	Sibling of case 7	Entire duodenum to rectum
12. Present case	F	none	Distal stomach to rectum

In the following twenty-eight years, only ten additional cases have been reported (1, 5-11), six with

Case Report

A 9 lb, 4 oz female child was born to a 37-year-old gravida III, para I Chinese woman following a normal spontaneous delivery. The mother had had one previous spontaneous abortion and had delivered a normal male child four years earlier. There

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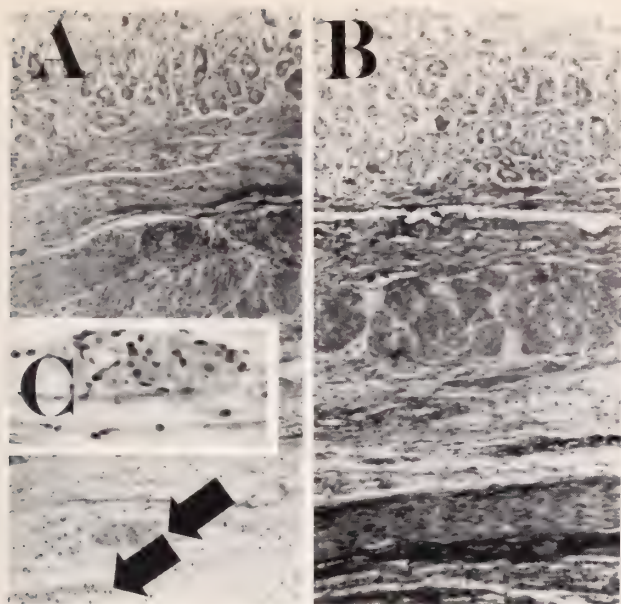


FIG. 1. Photomicrograph of stomach. A. Proximal stomach showing ganglion cells (arrows). Inset C is a high power view of a ganglion cell. B. Distal stomach; no ganglion cells are present.

was no significant prenatal history and no medications were given. The baby's Apgar score was ten at one minute, and ten at five minutes. Twenty-four hours after birth, the child began to vomit green fluid and had not passed meconium. X-rays were performed at three days of age, that showed dilated loops of bowel and air-fluid levels. A barium enema showed evidence of small bowel obstruction, and an exploratory laparotomy was carried out at five days of age. At surgery, the distal ileum and colon were collapsed, and there was dilatation of the mid-ileum. No definitive procedure was performed. Following surgery, the baby again became distended and vomited. An x-ray of the abdomen demonstrated residual contrast material from the previous barium enema. The infant was treated with intravenous fluids and at ten days of age was transferred to Beth Israel Medical Center. A rectal biopsy was performed, and was consistent with Hirschsprung's disease. A laparotomy was carried out at fourteen days of age. There was no evidence of dilatation of any part of the bowel. A biopsy of the right transverse colon was done, and no ganglion cells were seen. Frozen section studies of the transverse and ascending colon, cecum, appendix and multiple areas of the ileum and jejunum were carried out, and no ganglion cells were present. A biopsy of the jejunum at a point of 165 cm from the ileocecal valve was done, and no ganglion cells were seen. A

jejunostomy was performed at this point (22 cm from the ligament of Treitz) in the desperate hope that the obstruction from the aganglionosis would counterbalance the short length of bowel proximal to the jejunostomy.

Postoperatively, the infant continued to be obstructed. It soon became obvious that the child could not survive and after consulting with the parents, all extraordinary means of support were discontinued. She died of sepsis and peritonitis on the seventh postoperative day.

Postmortem examination revealed aganglionosis of the colon, small bowel and distal stomach. Ganglion cells were present in the esophagus and proximal stomach (Fig.1 A, B).

Discussion

Total intestinal aganglionosis is a rare, and invariably fatal disease. It is of interest to note that there were three pairs of siblings in the twelve reported cases and one other infant whose sibling had died of intestinal obstruction, the nature of which was not determined (10). For this reason, MacKinnon (11) believes that total intestinal aganglionosis may be a distinct entity, inherited as an autosomal recessive.

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Seminars in Physiology

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Fluid and Electrolyte Transport in Corneal Transparency*

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Introduction

The almost spherical eyeball is limited externally by a fibrous coat, the cornea and the sclera. They form a protective tunic and also withstand the intraocular pressure of about 15 mm of Hg. The anterior-posterior diameter of the eye varies in different species from 3 mm to 5 cm. In man, the diameter is 24 mm; and in the bull frog (the experimental animal we most commonly use), it is 12 mm. The curvature of the cornea is less than that of the sclera. Their junction forms a circular depression, the corneal sulcus.

The cornea is the transparent membrane that limits and forms the anterior wall of the eyeball. It has the shape of a convexo-concave meniscus. Its anterior side is in contact with the tear film and the air. Its posterior side is bathed by the aqueous humor of the anterior chamber, a fluid with almost the same refractive index as the cornea. Because of its limiting media (air and liquid), and its curvature, the cornea functions as a powerful convergent optical system with larger focusing power than the crystalline lens. It should be noted that if the lens was situated where the cornea is (between the air and the aqueous humor) and the cornea positioned between the aqueous and the vitreous humors, the lens would be by far the most powerful convergent

element of the eye. Thus, the cornea owes its remarkable focusing power partially to its location as the most anterior tunic of the eye.

Clearly, the most striking property of the cornea is its transparency which permits about 90% of incident light to be transmitted to the other internal components of the eye and eventually to the light receptors in the retina. In this review, we will deal with the remarkable anatomic structure of the cornea and with the physiologic mechanisms that control the hydration and transparency of the cornea. The effect of several pharmacologic agents on the regulation of corneal hydration will be reviewed.

Structure of the Cornea

The cornea consists of three parallel layers, the epithelium, the stroma, and the endothelium (Fig. 1). The total thickness of the human cornea is 500 μ (400 μ in the rabbit and 200 μ in the frog). The epithelium is a multicellular layer consisting of basal cells, intermediary cells, and the most superficial squamous cells. There are desmosomes connecting the basal and intermediary cells with tight junctions occurring in the most superficial cell layer. The electrophysiologic properties of the epithelium are similar to those of other tight epithelia such as frog skin.

The external surface of the epithelium has a fine ridge-type structure that seems to be important in keeping the tear film on the cornea surface. The most internal membrane of the cornea, the endothelium, is a unicellular layer that does not reproduce itself. Any endothelial cell loss is made up partially by enlargement of adjacent endothelial

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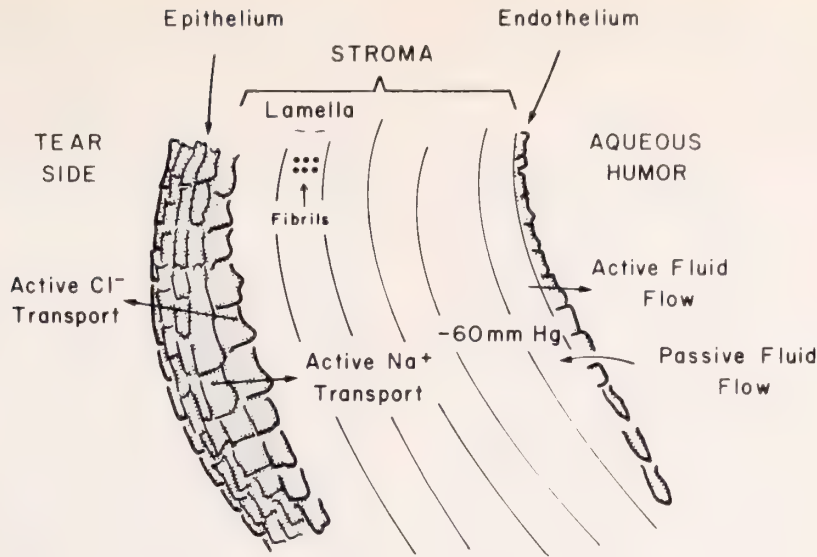


Fig. 1. Schematic representation of the cornea and its layers in the anterior to posterior direction.

cells. The endothelium belongs to the class of leaky (low electrical resistance and high permeability) epithelia such as the gallbladder and the proximal nephron of the kidney.

Between the epithelium and endothelium lies the stroma. This layer comprises about 90% of the cornea thickness. The stroma is made up of many sheets (about 200) of a fibrous material called lamellae, lying one on top of another parallel to each other as pages of a book. The main component of the lamellae are collagen fibrils, which are about 200 Å in diameter, and lie parallel to the corneal surface. The direction of all fibrils in a given lamella is the same, but the direction changes by an angle of about 90° for the fibrils of contiguous lamella. There are no fibrils situated in the anterior-posterior direction except in the stroma of some fish. Flattened cells, called keratocytes (modified fibroblasts), are mainly found between the layers of lamellae and occasionally within a lamella.

The fibrils in the lamellae are disposed with a remarkable regularity in a lattice-type arrangement. The spacing between fibrils is about 600 Å. The space between the fibrils is filled with a fluid substance containing about 4% mucopolysaccharides.

Just below the epithelium, there is a condensation of the stroma that forms a 12 μ thick membrane called Bowman's zone. Between the stroma and endothelium there is a basement membrane known as Descemet's membrane.

Transparency of the Cornea

Two main theories have been put forward to explain the transparency of the corneal stroma: 1)

the uniform refractive index and 2) the interference theory. When a beam of light travels through a medium it interacts with the matter in the medium. When it passes to another medium with a different refractive index it can, depending upon the incident angle, be reflected (does not penetrate the second medium) or refracted (it changes direction in the second medium). The refractive index of the corneal stroma is not uniform, as originally thought. The collagen fibrils have a refractive index of 1.55 (as does dry collagen) and the ground substance of 1.35. Thus, the uniform refractive index theory as a basis for corneal transparency can not be supported. In 1957, Maurice proposed the uniform size of the collagen fibrils and the near perfect regularity of the spacing between them as the basis of corneal transparency (1). When light travels through the stroma, it is refracted or scattered at the ground substance-fibrils interface. Because of the regularity of the collagen fibrils, the scattering by each individual fibril is not independent of the others. On the contrary, the destructive interference of the scattered light and the summation of all scattered beams results in zero net scattering to the side and complete transmission perpendicular to the surface plane of the cornea. It was later proven that a perfect lattice periodicity is not necessary. Benedek (2, 3) showed that even if the medium contains periodic fluctuations, there will be no scattering, provided that the fluctuations have a length smaller than half the light wavelength. According to this hypothesis, corneas with marked fluctuations in the periodicity of the collagen fibrils, such that the regions devoid of fibrils are as large or larger than the wavelength of light, should be opaque. This idea

has been substantiated by the observation of regions known as "lakes," devoid of fibrils, in opaque corneas.

Swelling and Opacification of the Cornea

When the cornea is isolated and placed in water or in Ringer's solution, it swells and loses transparency. The loss of transparency is caused by the absorption of fluid and the formation of "lakes" between the collagen fibrils. The swelling occurs only in the anterior-posterior direction and can continue for several days as the thickness of the tissue increases up to ten times its normal value. The composition of the bathing solution influences the rate of swelling, and corneas from different species swell to a different extent. Swelling will also occur in vivo if the epithelium or endothelium are damaged or destroyed (4). Swelling can be prevented in vitro by applying pressure with a plunger perpendicular to the stromal surface (5, 6). Swelling can also be prevented by immersing the cornea in a hyperosmotic solution of a non-diffusible solute (7).

In the normal cornea there is a force in the stroma that drives fluid into it and separates the corneal surfaces from each other. This swelling tendency is measured as a negative pressure inside the stroma and is called the imbibition pressure (8, 9).

The external pressure required to be applied on the corneal surfaces to prevent swelling is called the swelling pressure. It is opposite in sign but nearly equal to the imbibition pressure in vitro. In vivo, these two pressures differ by the value of the intraocular pressure. The value of the imbibition pressure has been determined to be about -60 mm Hg in a cornea with normal thickness (6, 8). As the stroma swells and its thickness increases, the negative pressure drops. Swelling stops when the pressure is zero. It is obvious that the intact cornea, in vivo, must possess a mechanism that prevents the stroma from swelling and counteracts the -60 mm Hg stromal pressure. This mechanism requires energy to operate and consequently must be coupled to metabolism and must be located in one or both cellular layers of the cornea: endothelium and epithelium.

The in vivo cornea is at a steady state that maintains its normal thickness (within narrow limits) in spite of the considerable negative pressure in the stroma. The negative pressure that draws fluid into the stroma across the endothelium and the epithelium must be exactly balanced by an equal fluid flow out of the stroma. It should be noted that either the fluxes or the forces should be balanced.

In the latter case, a concentration gradient of about 3.3 mOsm between the stroma and the bathing media is required to oppose a pressure of 60 mm Hg (assuming solute impermeability of the limiting barriers). To understand how this balance can be achieved, it is necessary to know not only the active mechanisms operating at these barriers but also their passive properties.

Control of corneal hydration as associated with metabolism was first demonstrated by Davson (10) and by Harris and Nordquist (11) who found that the cornea of an enucleated eye will swell faster if metabolism is depressed by lowering the temperature to 10° C. By rewarming the eye to 37° C, the cornea thins as fluid is pumped out of the stroma. The importance of the endothelium and epithelium, as both, barriers to the movement of solutes and fluid and as sites of active transport of electrolytes and fluid, has been clearly demonstrated in the last twelve years, and will be discussed below.

Fluid Transport by the Corneal Endothelium

In this section, we will be concerned with passive permeability of and active fluid transport by the endothelium. In general, the permeability properties of a simple membrane can be completely described by three independent coefficients: hydraulic conductivity L_p ; solute permeability ω ; and the reflection coefficient σ .

L_p for the rabbit endothelium has been determined as 1.55×10^{-11} cm³ sec⁻¹ dyne⁻¹ (12). Since a 60 mm Hg pressure is equal to about 79×10^3 dyne/cm², the imbibition pressure of the stroma should cause a flow of water across the endothelium of about 4.3 μ l/hr·cm² ($J_v = L_p \cdot IP$; J_v is the volume flow and IP the imbibition pressure of the stroma). This simple relationship is only true for simple membranes in the absence of a solute gradient. Thus, it is remarkable that experimentally measured flows are in the range of 3 to 12 μ l/hr·cm² (13-19).

Primary water pumps have not been described in any biological membrane. Thus, active water or fluid transport is always secondary to active solute transport. The manner in which active solute transport prevents stromal swelling may be explained in two basic ways. One possibility is that the solute transport creates a concentration gradient that exactly opposes the imbibition pressure ($IP = \sigma RT \Delta C$). In this case, the forces are balanced and there is no net flow across the membrane. A second possibility is that the concentration gradient actually produces a net fluid flow, but because of the heterogeneity of the membrane a backleak occurs

across a parallel pathway; that is, two measurable fluxes occur across the membrane but the net flow is zero.

The single endothelial cell layer is actually a heterogeneous structure that can be viewed simply as two membranes in series (the aqueous side and the stromal side cell membranes) which are in parallel with the paracellular pathways. Because of this, most measurements of endothelial permeability denote a combination of the characteristics of the various elements involved. In the isolated swollen cornea when the IP is zero and the passive forces are eliminated, the active mechanism produces a net fluid flow of about 3 to 6 $\mu\text{l/hr}\cdot\text{cm}^2$ in the stroma to aqueous direction (19, 20).

Direct evidence that the integrity of the endothelium was necessary to maintain normal thickness in the isolated rabbit cornea was provided first by Mishima and Kudo (21). They were able to maintain normal thickness for ten hours by perfusing the endothelium with a solution containing glucose, vitamins, cofactors and amino acids. They were also able to thin a cornea that was preswollen at 4° C by perfusing it with the solution at 37° C.

In 1972, Dikstein and Maurice (13) and Maurice (14) described a technique by which an isolated cornea can be perfused and its stromal thickness determined. Using this technique, they were able to show that the endothelium was capable of pumping fluid out of the stroma against a head of pressure and that sodium, bicarbonate, oxygen, adenosine and glutathione were necessary for optimum operation of this fluid pumping mechanism.

The work of Fischbarg et al (15, 22), Hodson (18), and Barfort and Maurice (19) showed that a small electrical potential difference (PD) of about 0.5 mV can be measured across the corneal endothelium. Inhibiting the PD (with ouabain or by omitting sodium from the medium) also inhibited the fluid pump. However, carbonic anhydrase inhibitors were effective inhibitors of the fluid pump but not of the PD (15). Another disturbing finding was the orientation of the PD; the stromal side was positive. This indicates that if Na^+ was the transported solute, it was transported in the wrong direction. To explain this anomaly, Fischbarg and Lim (15) and Hodson (18) proposed models in which a coupled mechanism will pump a neutral salt into the aqueous humor creating the concentration gradient to move fluid. The PD, they suggested, was simply created by ionic diffusion across the intercellular channels. Later, Hodson et al abandoned this theory and proposed in a series of articles (20, 23–25) that both PD and fluid pumping can be explained

by the active transport of bicarbonate from the stroma to the aqueous humor. One weakness in this theory is that the experimental data is also consistent with a proton pump in the aqueous to stroma direction. A similar controversy regarding the distinction between bicarbonate and proton transport exists in the turtle bladder preparation (26, 27).

A different approach to explain the fluid pump of the corneal endothelium has been taken by Shapiro and Candia (28) and by Rehm and Spangler (29). Their models are similar to that of Curran (30) and Durbin (31) to explain the apparent active transport of water across the intestine. The bases of the model are: 1) different solute reflection coefficients on the stromal side and aqueous side of the endothelial cell membranes; and 2) an osmolarity difference between the cell compartment and the surrounding medium. This could be maintained by the metabolic breakdown of solutes or by nonspecific pumps. The main feature of this model is fluid recirculation. Fluid is actively transported across the transcellular pathway towards the aqueous humor. If flows back passively towards the stroma via the paracellular pathway. This circulation will favor nutrition from the aqueous humor and waste removal from the avascular cornea.

Ionic and Fluid Transport by the Corneal Epithelium

The corneal epithelium belongs to the class of tight epithelia such as frog skin and toad bladder. Its solute permeability is much lower than that of the endothelium. From evaporation, the salt concentration in the tear film is much larger than in the stroma. This causes a solute concentration gradient, which is partially maintained by the low solute permeability of the epithelium. The gradient produces a fluid flow of about 3 $\mu\text{l/hr}$ which balances an equal amount of water evaporated when the eye is open (32). It has been found that 3 $\mu\text{l/hr}$ move from the aqueous to the tears, although the amount of water entering the stroma is equal to the amount leaving it. It is logical to infer that when the eyes are open, the cornea should be slightly thinner, and the stromal IP more negative to account for the 3 $\mu\text{l/hr}$ net flow into the stroma across the endothelium. Obviously, when the eyes are closed, there is no fluid transport across the stroma, only fluid recirculation across the endothelium.

Clearly the epithelium plays an important, albeit passive, role in controlling corneal hydration and transparency. There is, in addition, some evidence indicating that the epithelium may also play an active role. In 1959, Donn et al (33) showed that

the isolated rabbit corneal epithelium maintains a PD of about 15 mV, the stromal side positive. This electrical activity was due to a net active Na^+ transport towards the stroma. The direction of this transport was opposite to that required to move water out of the stroma. In 1966, Zadunaisky found that the isolated frog corneal epithelium also maintained a PD of about 20 mV, the stromal side positive (34). In this case, however, the electrical activity was caused by an active Cl transport from the stroma to the tears. This finding was important evidence in favor of active fluid transport out of the stroma by the epithelium. The fact that the rabbit and frog corneal epithelia displayed transport properties so different was further examined by several investigators. The work of Candia and Askew (35), Candia et al (36) in the frog and that of Klyce et al (37) in the rabbit, demonstrated a sodium pump in the frog cornea and a Cl pump in the rabbit cornea respectively, indicating that corneas from both species did have both transport systems. The Cl pump can be stimulated with cAMP (37), epinephrine (38, 39), theophylline (37), ascorbate (40), and the calcium ionophore A23187 (41). It is conceivable that in vivo the Cl pump is under a continuous state of stimulation due to the presence of cAMP and a high concentration of ascorbic acid in the aqueous humor.

The importance of these active transport systems in the corneal epithelium for the maintenance of transparency was demonstrated by Klyce who found that the stimulated rabbit corneal epithelium is capable of thinning a swollen cornea (42). Candia has presented similar evidence working with the frog cornea (43).

In addition to its importance to corneal physiology, Cl transport in the cornea has an added interest to physiologists in general. It resembles Cl transport in the thick ascending limb of Henle's loop. It is Na^+ dependent (34, 44, 45) and inhibitable by ouabain (44). The diuretics, ethacrynic acid, furosemide and bumetanide are also potent inhibitors of Cl transport in the cornea (46, 47).

It is now apparent that active transport processes in the endothelium as well as the epithelium contribute to the maintenance of corneal transparency.

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Student's Corner

Paul Celano, B.S., Editor

The "On-Off" Phenomena In Parkinson's Disease—Treatment and Theoretical Considerations

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Abstract

The "on-off" phenomenon, one of the limiting factors in the use of L-Dopa in the treatment of parkinsonism, occurs in approximately half of the patients using the agent for five years or more. The effect of Deprenyl, a monoamine oxidase-B inhibitor, added to an existing regimen of L-Dopa/Carbidopa on this phenomenon was evaluated. The study was carried out in a double blind crossover fashion and preliminary results indicate: a reduction in "on-off" phenomena, a more effective control of parkinsonism symptoms, but an increased appearance of dyskinesias. The implication of these findings in relation to current concepts regarding the striatal effects of these pharmacological agents is discussed.

Introduction

Parkinson's disease is an incapacitating neurologic disorder of unknown etiology characterized by increasing levels of disability and a high rate of coincident mortality (1). As knowledge of the morphological and biochemical changes underlying the disorder has increased, it has become evident that the parkinson syndrome is related to a depletion of striatal dopamine (2, 3). This led to the development of the use of L-Dopa in the treatment of parkinsonism and heralded a new era in the therapy of the disease. The initial clinical response to L-Dopa was found to be somewhat variable (4-6), but was generally positive and dramatic (2, 5, 7, 8). Long-term use of the agent has resulted in a more prolonged period of reduced symptomatology, and

a reduction of the increased mortality seen with the disease (8, 9). Unfortunately, prolonged L-Dopa use results in a decrease in therapeutic response (8-12) with a concomitant increase in side effects (9, 12). One of the most distressing of these side effects is the so-called "on-off" phenomenon.

A phenomenon related to "on-off," the Akinesia paradoxa, was recognized as a classical symptom of Parkinson's disease prior to the introduction of levodopa (11, 13). Patients in an akinetic state could be suddenly activated for short periods of time by sensory stimulation (11). This abrupt change in activity bears striking resemblance to the L-Dopa induced "on-off" reaction that is defined as a sudden onset of akinesia followed by an abrupt return of mobility that may be accompanied by dyskinesia (4). The "on-off" phenomenon develops in approximately 50% of parkinsonian patients who have been treated with L-Dopa for five years (10, 14). Affected individuals are described as having a lower mean age at the onset of disease (10, 14), and a higher maintenance dose of levodopa (10) as compared with patients who do not exhibit the phenomenon. Patients with "on-off" alternate between

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being asymptomatic or dyskinetic (on), and having frank parkinson symptoms (off) (15). Two different forms of "on-off" have been described (11, 13, 16). The first depends on the timing of doses of L-Dopa, while the second is not dose related. Early morning akinesia, start dose akinesia, peak dose dyskinesia, end dose akinesia, and diphasic dyskinesia are related to the former while the latter occur randomly.

The occurrence of "on-off" has been correlated with plasma L-Dopa levels in some, but not all cases (13, 14, 16, 17). High plasma L-Dopa concentrations correlate with asymptomatic or dyskinetic periods, while low levels are found when patients are parkinsonian. These variations in plasma L-Dopa concentrations have been interpreted as being reflective of similar changes in the level of the agent in the brain. As a result, therapeutic efforts were aimed at minimizing fluctuations of plasma L-Dopa concentrations. Frequent small doses of L-Dopa, low protein diets (13, 17), the use of a peripheral decarboxylase inhibitor (12, 18), and constant intravenous (IV) infusion of L-Dopa (17) have had only a limited success in alleviating the "on-off" reaction. More recently, attention has been directed to ways and means of altering brain dopamine itself.

Monoamine oxidase (MAO) is a critical enzyme in the metabolic pathway involved in the breakdown of dopamine to homovanillic acid (12, 19, 21). Inhibiting MAO offers the potential of extending the effect of a dose of L-Dopa by protecting brain dopamine from degradation. Until recently, agents capable of such action have been contraindicated in combination with L-Dopa as they were found to produce hypertensive crises. More recent work has shown that MAO exists in two forms (types A, B) which have differing substrate specificities. Hence, differing reactions are expected to result from selective inhibitors of each. In the human brain, MAO-B is responsible for dopamine metabolism. An agent that acts selectively to block MAO-B would have the desired effect on brain dopamine levels, but would avoid the side effects that result from the inhibition of MAO-A. The present study investigates the effects of a selective MAO-B inhibitor, Deprenyl (phenyl-isopropyl-methyl propynylamine hydrochloride) (19, 20, 24) on parkinson patients who exhibit the "on-off" phenomenon.

Materials and Methods

Five patients taking part in a double blind cross-over trial of Deprenyl, a monoamine oxidase-B inhibitor, were observed. All candidates for the study

exhibited the "on-off" phenomenon and gave informed consent after the nature of the procedures had been explained. Patient profiles are presented in Table I.

Patients were admitted to the clinical research center at The Mount Sinai Hospital for a 28-day stay. During the first six days, patients were stabilized on a reduced maintenance dose of Sinemet (L-Dopa + Carbidopa). Deprenyl (5 mg b.i.d.) or a placebo was then administered with the reduced dose of Sinemet for ten days. The patients were then crossed over from Deprenyl to placebo or vice versa, and this regimen continued for a period of twelve days. Some adjustments of Sinemet dose necessitated by the patients' clinical state were made during the study. When made, these changes involved the dose and not the timing of doses.

Hourly rated neurologic examinations were performed by nurses from 0700 to 1900 hours. Personnel restrictions did not allow for the rating of each patient for every day of his stay. A score of +12 indicated maximal dyskinesias, while -15 indicated maximal parkinsonism. A score of 0 indicated a normal exam. In addition, the investigator performed a more detailed rated neurologic examination (scale of +48 to -100) on three of the patients on a three to five times a day basis.

The results of each day's rated neurologic examinations performed by the nurses were averaged for each patient. From these data, mean values for parkinsonism and dyskinesia were calculated for control (placebo + last three days of initial six-day period) and treatment (Deprenyl) periods. In addition, the number of "on-off" reversals were calculated for each period. The patient was considered "off" whenever parkinsonism was present as indicated by the nurses' rated neurologic examination. A reversal consisted of a change from parkinsonism to normal or dyskinesia (or vice versa) on the next hourly exam. The significance of differences between control and treatment periods were calculated using Student's t-test.

TABLE I
Patient Population

Patient	Sex	Age, Onset Parkinson's Disease	Present Age	Years on L-Dopa	Parkinson's Disease Stage*
E.K.	F	56	66	6	III
E.Z.	F	50	65	11	III
J.L.	M	39	49	4	II
D.S.	F	57	75	9	III
C.S.	F	46	54	6	III

* Stages based on that described by Hoehn and Yahr (1)

Results

Data on one patient (E.K.) are not included in the analyses performed. She was hypertensive at the start of the study and began to receive alpha-methyl-dopa during the course of her stay.

Data on the levels of parkinsonism, dyskinesia, and the number of "on-off" reversals are presented in Table II for control and treatment periods. Three out of four patients had a statistically significant decrease in their parkinsonism during the treatment period ($p < .01$). Although there was a trend towards increased dyskinesia in treatment vs. control period, the trend was not statistically significant. Two of four patients had a statistically significant decrease in the number of "on-off" reversals in treatment vs. control periods. In the other two, nonsignificant increases were observed. Based on average data for all patients, during treatment periods, patients show a decrease in parkinsonism ($p < .01$), an increase in dyskinesia (NS), and a decrease in the number of "on-off" reversals (NS).

C.S. exhibited parkinsonism and dyskinesia si-

TABLE II
Results of Deprenyl Trial

Patient	Control			Treatment			t
	$\bar{x}$	s^2	n	$\bar{x}$	s^2	n	
<i>Parkinsonism</i> (absolute value of score)							
E. Z.	4.87	1.25	13	2.78	2.32	12	3.94**
J. L.	1.76	0.16	12	0.88	0.09	8	5.35**
D. S.	1.08	0.19	12	1.09	0.18	10	0.05
C. S.	3.01	0.37	9	1.76	0.45	8	4.01**
All Pt	2.71	2.78	46	1.72	1.51	38	3.04**
<i>Dyskinesias</i>							
E. Z.	2.95	1.29	13	3.75	0.98	12	1.87
J. L.	1.10	0.24	12	1.38	0.25	8	1.25
D. S.	0.41	0.11	12	0.38	0.08	10	0.22
C. S.	1.64	0.19	9	2.06	1.00	8	1.14
All Pt	1.55	1.44	46	2.14	2.78	38	1.88
<i>On-Off Reversals</i>							
E. Z.	5.69	3.91	13	3.92	1.41	12	2.68*
J. L.	5.33	0.39	12	3.75	2.19	8	3.14**
D. S.	4.83	2.47	12	5.50	2.45	10	1.00
C. S.	2.78	0.84	9	3.13	1.61	8	0.68
All Pt	4.80	3.11	46	4.07	2.86	38	1.92

** $p < .01$

* $p < .05$

The results of each day's rated neurologic exams performed by the nurses were averaged for each patient. From these data, the number of "on-off" reversals, the level of dyskinesia, and the level of parkinsonism were calculated for control (placebo + last 3 days of initial 6 day period) and treatment (Deprenyl) periods. A score of 12 indicates maximal dyskinesia while 15 indicates maximal parkinsonism (absolute value of parkinson score). A score of 0 indicates normal exam. "On-off" reversal is a change from parkinsonism to normal or dyskinetic (or visa versa). The significance of differences between control and treatment periods were calculated using Students t-test.

Average results for all patients were calculated (All Pt).

multaneously during several examinations performed in control and treatment periods. The phenomenon was, however, seen mainly towards the end of the Deprenyl treatment period and occurred mainly in the afternoon. At these times, the patient's overall clinical state was at its best (absolute score closest to 0). The data in Table III are derived from the investigator's rated neurologic examination performed during one of these periods.

Discussion

The results of this study, while tentative, support other work (19, 20, 22) and indicate that Deprenyl

TABLE III
Rated Neurologic Examination of Patient C. S.

Score	Description of Rating
<i>Parkinsonism Ratings:</i>	
Rigidity:	
RUE	2 mild to moderate
LUE	1 slight or detectable only when activated by other movements
Neck	2 mild to moderate
RLE	0 Absent
LLE	2 mild to moderate
Finger tap: (pt. taps thumb with forefinger, then with each finger in rapid succession)	
RIGHT	1 slightly slow
LEFT	1 slightly slow
Succession movements: (pt. taps knee alternatively with palm and dorsum of hands)	
RIGHT	1 slightly slow
LEFT	1 slightly slow
Foot tap: (using heel as fulcrum, pt. taps floor with ball of foot as rapidly as possible)	
RIGHT	4 unable
LEFT	2 slow
Arising from chair:	
	2 pushes self up from arms or seat
Standing posture:	
	1 not quite erect, slightly stooped posture
Bradykinesia:	
	1 minimal slowness giving movements a deliberate character
<i>Dyskinesia Ratings:</i>	
RUE	2 mild
LUE	0 absent
RLE	2 mild
LLE	1 minimal
Shoulder	1 minimal
Pelvis	1 minimal

RUE—Right upper extremity

LUE—Left upper extremity

RLE—Right lower extremity

LLE—Left lower extremity

Significant results of a rated neurological examination performed by the investigator (L.G.) on patient C.S. Scale for each parameter is 0-4. Examination results are consistent with the occurrence of simultaneous parkinsonism and dyskinesia in this patient.

potentiates the effects of L-Dopa resulting in decreased parkinsonism, increased dyskinesia, and a decreased frequency of "on-off" reversals. Treatment with Deprenyl, however, does not abolish "on-off" fluctuations in all patients or all the time in patients with a response (19, 20, 22).

The therapeutic effect of Deprenyl must be related to its mode of action. As an MAO-B inhibitor, it acts to block the intraneuronal breakdown of dopamine (19, 20). This may produce higher, more sustained synaptic levels of the agent. The ergoline, bromocriptine, has a prolonged, selective dopaminergic receptor action (23). It too, results in an increase of dyskinesia, a decrease of parkinsonism, and a reduction in fluctuations in clinical state (23).

Other attempts have been made to provide constant levels of L-Dopa in an attempt to reduce "on-off" episodes. Constant IV administration of L-Dopa results in an leveling of clinical response; however, "on-off" returns with movement or stress (17). Low protein diets aimed at limiting the competition for absorption of L-Dopa from the gut has limited benefit, as does the administration of small doses of L-Dopa at frequent intervals (14). Thus, the strict control of L-Dopa levels may modulate the "on-off" effect, but does not result in its abolition. Further, we have demonstrated that parkinsonism and dyskinesia can exist simultaneously in a patient (Table III). These observations indicate that more than one factor may be involved in the control of the "on-off" phenomenon.

Two types of dopaminergic receptors have been demonstrated in the rat (24). One is found on dopaminergic neurons and is primarily presynaptic in nature. It is adenylate cyclase independent and it binds apomorphine preferentially to haloperidol and spiroperidol. The second is found in postsynaptic, nondopaminergic neurons located in the striatum. It is adenylate cyclase dependent and binds haloperidol and spiroperidol preferentially to apomorphine.

Both pre and postsynaptic receptors bind apomorphine, but in a dissimilar manner. Elimination of higher affinity presynaptic receptors could result in an increase in the relative representation of lower affinity postsynaptic receptor sites (24). Nigro striatal pathway lesions were found to produce a decrease in apomorphine binding in the striatum (24), presumably reflecting the loss of presynaptic receptors, while simultaneously potentiating behavioral responses to apomorphine, which are presumably due to postsynaptic activities. The supersensitivity has two components. There is an acute response caused by presynaptic denervation, and a chronic, gradually developing one due to increased response

at the postsynaptic level (26). One implication of this evidence is that degeneration of the nigrostriatal pathway could result in a progressive alteration in the populations of two sets of dopaminergic receptors. These changes may have behavioral counterparts.

In humans, the long-term treatment of schizophrenics with neuroleptic agents results in an increased binding of ^3H haloperidol in the striatum. This may reflect a proliferation of postsynaptic dopaminergic receptors resulting in a supersensitivity that could underlie the tardive dyskinesia seen in some of these patients (27).

An increase in ^3H haloperidol binding has also been reported as occurring in the putamen of untreated parkinson patients (28). The predicted decrease in ^3H apomorphine binding is also found. Administration of apomorphine has been reported to reduce the dyskinesia produced by L-Dopa (29, 30). If the agent is given during periods when the effectiveness of L-Dopa is decreasing, it can result in the abolition of the "on-off" phenomena (22).

It may be the relative effect of dopamine receptors which bind haloperidol with respect to those that bind apomorphine which is of importance in "on-off" phenomena. Since treatment with L-Dopa results in normal ^3H haloperidol binding (28), the development of "on-off" may be related to relative changes in the apomorphine binding receptors. Stimulation of apomorphine binding dopaminergic receptors may result in the abolition of dyskinesia, while action of the haloperidol binding dopaminergic receptors may produce an alleviation of parkinson symptoms.

Conclusions

Treatment with Deprenyl can result in decreased parkinsonism and increased dyskinesia by protecting brain dopamine from degradation. Thus, through the use of Deprenyl, a positive therapeutic response may be achieved with lower dosages of L-Dopa. This is, however, a preliminary result based on a small number of cases. A larger study will be necessary before definitive results are obtained.

The co-administration of L-Dopa and Deprenyl may result in decreased, but not abolished "on-off" reversals. In the past, therapeutic regimens that were designed to produce more constant serum L-Dopa levels have had only a limited success in alleviating the "on-off" reaction. In some cases, "on-off" phenomena may be related to changes in serum L-Dopa levels while in other cases, fluctuations in clinical state occur in the face of constant serum levels of the agent. Further, parkinsonism and dys-

kinesia may occur simultaneously in a patient. One possible explanation of these findings is that at least two factors may be involved in the occurrence of "on-off" reactions, such as changes in brain dopamine levels and fluctuations in the relative sensitivities of two sets of dopaminergic receptors. Deprenyl may act to reduce "on-off" reversals by providing more sustained brain dopamine levels, thereby affecting the first of these two factors. Administration of the agent may not result in the abolition of "on-off" phenomena because of changes in the sensitivities of two sets of dopaminergic receptors.

At the present time, the absence of positive evidence in support of the two-receptor hypothesis requires that its tentative nature be stressed. If the hypothesis holds, attempts to abolish "on-off" phenomena by manipulation of brain dopamine levels alone may be unsuccessful in many cases. Therapeutic regimens would have to be designed that would take into account both brain dopamine levels and differential receptor sensitivities.

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ANNOUNCEMENT

The first **S. B. Gusberg Seminar on Reproductive Immunology** will be held on June 26 and 27, 1980 at The Mount Sinai Medical Center, One Gustave L. Levy Place, New York, N. Y. 10029.

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Presenting authors of accepted abstracts will be exempt from the registration fee. Proceedings of this seminar will be published in the form of a Festschrift by The Mount Sinai Journal of Medicine. Instructions to authors will be mailed together with the notice of acceptance for submitted abstracts. Reports at the meeting will be limited to a 10 minute presentation followed by a 5 minute discussion. For further information write to: Dr. N. Gleicher or call (212) 650-6555.

Erratum

Please note: The correct title for the lead article in September/October 1979 (Vol. 46, No. 5, pp. 431-454) by Drs. Marshak, Lindner, and Maklansky should read:

Crohn's Disease of the Colon (Granulomatous Colitis)

Additionally, the following Table was inadvertently omitted.

Crohn's Disease of the Colon (Granulomatous Colitis)

TABLE
Roentgen Findings in Granulomatous Colitis

-
1. Tiny discrete ulcers (aphthoid ulcers) usually associated with small, radiolucent nodules.
 2. Segmental distribution, more disease on right than on left.
 3. Universal distribution in colon almost always associated with involvement of terminal ileum.
 4. Nodular, irregular pattern after evacuation.
 5. Transverse ulcerations.
 6. Longitudinal ulcerations.
 7. Combination of transverse and longitudinal ulcerations with cobblestoning.
 8. Strictures.
 9. Irregularity of contour with asymmetric involvement and pseudodiverticula.
 10. Skip lesions.
 11. Fistulas, frequent in ileum, considerably less common in colon.
 12. Intramural abscesses.
-

The Long-term Care Challenge: Rationalizing a Continuum of Care for Chronically Impaired Elderly

ROSE DOBROF, D.S.W., JONATHAN M. METSCH, Dr. P.H., HARRY R. MOODY, Ph.D.,
THOMAS C. CHALMERS, M.D., AND PREMA MATHAI-DAVIS, Ed.D.

Introduction

Case: The hospital utilization review committee rules that the 82-year old, poststroke patient no longer needs or is eligible for hospital care. She can't go home alone. The services she needs cannot be pasted together as quickly as is required, or with the confidence that they will remain in place as long as is needed. Small wonder, then, that there are inappropriate institutional placements. Older people are in health related facilities who could have made it at home or are in an enriched housing situation if community based services were available. Older people reside in skilled nursing facilities who could receive the care they need in a less restrictive setting. "But at least she'll be taken care of" agree the professionals and the family. Thus, the older person, more often than not, grieves for her lost home and life, but makes the best of it. She knows that everyone tried, and there really was no alternative.

Long-term care services for the chronically impaired elderly are characterized by fragmentation and discontinuity. Coinhabiting this fragmented world are the older people, their families, neighbors and friends, and the professionals who treat them—the family doctor, the nurse and social workers in the acute treatment hospital, the senior center director, and the visiting nurse. It is not that the will is not there. Although the most vulnerable of the

old are those without functional families and other informal supports, basically the majority of older people do have families, and the families want to do "what is best" for their aged members. The professionals share this goal, wanting to help the elderly secure the services needed to live out their remaining years in the least restrictive and most comfortable environment possible. Equally, the professional wants to help the family of the older person do what is best and behave in a responsible way. Fragmented, however, means incomplete. Sometimes the service is simply not in place. Sometimes it is there, but there is a waiting list. Or, because of income, health status, place of residence, lack of resources, knowledge, or skill in negotiating the system, the older person is not eligible or able to enter the service system effectively.

Fragmented also means disconnected. Social workers in hospitals report on their efforts to paste together the array of services available. The chronically infirm older person may need one agency for one service, another, for another to realize the wish to return home (for example, a visiting nurse, a home health aide, meals-on-wheels, a physician who will make house calls or transportation and escort service to the doctor's office or the outpatient department of the hospital). One agency is a Title XX service, and, therefore, income tested; another is Medicare funded, and may have limitations on the duration and range of services that bear no relation to the reality of chronicity. Additionally, Administration on Aging (AoA) programs with eligibility requirements differ from Social Security Act programs.

The reality of discontinuity reflects the lack of an operational continuum of care: that is, a system of

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identification and placement that assures, with a reasonable degree of certainty, that a chronically impaired individual's need for health and social services is met at the level appropriate to the extent of the incapacity, over an extended period of time, with the appropriate type and intensity of intervention.

This paper shall review the historical context that has shaped the nonsystem of care and shall define how a continuum of care can be rationalized with the objective of providing patient-centered, cost-effective, long-term care services. Finally, it shall report how a joint Mount Sinai School of Medicine/Hunter College planning effort has been initiated to address the issues raised.

Background

How do we account for the absence in most United States communities of a rational, comprehensive, and accessible system of health and social services for functionally disabled older people? First, there is the history of service development—the lumping together of dependent groups—the mentally ill, the children, the aged, the indigent, the handicapped, *inter alia*, without reasoning why the dependency or differential service requirements. A pervasive ambivalence exists about dependency and selfsufficiency that has sometimes been reflected in a reluctance to make public provisions, and in the niggardliness with which provisions are made. Our complex system, has responsibilities located at federal, state, and local governmental levels with a mix of public, voluntary, and proprietary auspices for service provisions. There is the ethnic heterogeneity of our population that often differentiates access and quality of services with ethnicity. Changes in programmatic approaches occur as new technology is created or as ideas and values evolve.

For example, there are identifiable periods of belief in the efficacy of institutional care (the 1820's, the post-Civil War period, the 1950's). Then, there are periods in which the institution is viewed as a last resort arrangement or reflecting the decline of the family or the paucity of other provisions. There are important inter- and intra-state differences. Level and range of provisions are often functions of per capita income. States vary with respect to the percent and absolute number of their aged citizens. There are differences associated with the urban, suburban, and rural composition of the state. Even the auspices of long-term care programs vary among the states: homes for aged under voluntary auspice are more numerous in the Eastern seaboard states

and less predominant in Southern and Western states. In poorer and less urban states, Arkansas, for example, there are almost no facilities under voluntary auspices.

The 1950's, 1960's and early 1970's were, for many states, an era of rapid expansion of institutional care of functionally disabled older people, with little attention, except in recent years, given to community based services. The push for de-institutionalization or for placement when necessary in the least restrictive facility is relatively recent (the last decade or so) and New York's experience has been shared by many other states. The absence of a comprehensive system of community services has made achieving the dream of de-institutionalization impossible.

Paradoxically, the affluence of the American economy and the richness of our resources have been obstacles in the development of a rational and comprehensive system of long-term care. In other countries, such as the Scandinavian nations, England, and Scotland, economic necessity prompted new services, linked together by mechanisms that guaranteed access to appropriate services with continuity of care, based on careful assessment of need.

Particularly in the decades following World War II, there was no such pressure in the United States. Hill-Burton and other federal and state funds were available for capital construction. The Medicare and Medicaid amendments to the Social Security Act made reimbursement monies available to health care agencies and professionals. Title XX of the Social Security Act provided new monies to states for social service programs; and the Older Americans Act, particularly after 1973, became the source of support for community based programs. Public officials and service providers recognized that, although new programs were being created and new facilities constructed, the absence of coordination at the macrolevel with linkage mechanisms at the direct service level meant that many functionally disabled older people were being denied the services they needed. It is only in the past few years that it has been recognized—despite the knowledge and technology presently available, and despite the excellence of many of the health and social service programs already in existence—the twin goals of effectiveness and efficiency cannot be met without rational planning and service coordination.

Recent Trends

It is estimated that public expenditures for long-term care amounted to almost six billion dollars in

fiscal 1975 and, at the same time that private funds were expended in at least this same amount. These expenditures represented increases far in excess of the rise in monies spent for all health care. Furthermore, projections for future years increase the spending for long-term care. The continuing growth of the 75+ cohort—the group in which the incidence of chronicity and disability is highest—is the major factor in accounting for the projections of increasing need for long-term care in the years to come. There are, however, other societal trends that require notation.

It is estimated that less than half of the disabled or impaired older people in need of health and social services receive these services under publicly funded programs. Some simply do not receive the care and services their declining health status warrants until, or unless, a crisis occurs. For many, the “hands on” care continues to be provided by family members and, to a lesser extent, by friends and neighbors. Despite the importance of the family as care givers, several long-term demographic trends are already compromising the ability of families to provide this care.

First, and perhaps of greatest significance, is the enormous increase of women in the labor force. Historically, the responsibility for “hands-on” care of infirmed older people has been the task of the women in the family. “A daughter’s a daughter all her life; a son’s a son until he gets a wife,” like many folk sayings, is a reasonably accurate summation of the real world. All studies on the family relationships of older people testify to the salience of the role of the female members of the family in the performance of both affective and instrumental tasks necessary to the care of the older family members.

These studies also testify to the continuing vitality of the American family and the strength of the ties of family devotion and responsibility to older members. Nonetheless, the presence of increasing numbers of women in the labor force and the likelihood that this trend will continue means that millions of women, who a generation ago would have provided the daily care that disabled or impaired older people require, are simply not available for this task. Studies indicate that the absence of day-time care giving relatives is, for many families, a critical factor in the decision to seek institutional care for a functionally disabled family member. Parenthetically, it should be noted that these studies were done in a metropolitan area where, as in many areas of the nation, there has been an enormous increase in long-term care facilities, but not a

similar increase in community based, long-term care services. In some cases, it seems clear that had the full panoply of community services been in place, many older people might have been able to remain in their own homes.

The impact of the entry of women in the labor force on child care has become a matter for research and public policy analysis, but little attention has been paid to its impact on long-term care of functionally disabled older people. Given all the forecasts of increasing numbers of women in the labor force and, therefore, their decreasing availability for daily care giving tasks, additional public provisions, both institutional and community based, are going to be required and must be taken into account in the service projections for the coming decades.

Second, the implications for service needs because of the growth in the 75+ age group is not simply a function of the high incidence of disability in this group. It must also be recognized that the children—the key care giving relatives—of the oldest cohort are themselves frequently facing the problems and crisis of the later years of the life cycle. A daughter in her 60’s, for example, may be the care giving relative for her mother in her 80’s. The retirement, illness, or death of her own husband may lead to a change in her living situation such as, a move away from her mother to a retirement community or a move to be closer to her own adult children. The consequence is the removal of the daily support that had enabled the mother to remain in her own home. In the absence of guaranteed public provision of the community services the mother needs in the decades to come, the advanced age of many of the care givers, and the consequent limitations on their service providing capabilities must be considered.

A third item that will have an impact on future public provisions is difficult to estimate, but merits consideration. This has to do with the expectations people have, and what they believe should be provided out of the public treasury. Despite the calls for tax cuts, for reductions in public expenditures at all levels of government, despite continued debate about the proper division of labor between the public and private sectors and the question about the percentage of the gross national product that can be allocated to health and social services, the basic service entitlements of the aged—particularly the poor and functionally disabled—are less often challenged than are the entitlements of other service needy groups. In New York State, for example, efforts to secure improved assistance levels, notably in the Aid to Families of Dependent Children pro-

gram fail regularly, while the Supplementary Security Income benefit level is adjusted upward. Efforts at cost containment, particularly in the area of long-term institutional care, are made and eligibility requirements are tightened to prevent inappropriate or unnecessary placements. The basic entitlement of older people to income maintenance and health and social services is not seriously challenged. Instead, the press is for more and better public provision. This demand by older people themselves, their families, and their advocates, for improvements in the quality and range of services seems likely to continue in the decades ahead. It is a force that must be taken into account in future service projections.

Collectively, these factors among others, clearly suggest that there will be increasing numbers of service needy elderly and continuing demand for public program development, if not public provision of these services. It is also selfevident that program development efforts must address, not compound, the issues of fragmentation and discontinuity.

Continuum of Care

Introducing the notion of a continuum of care suggests that the manner of health and social services for the chronically impaired elderly can be logically ordered and integrated. However, even the process of conceptualization of a continuum is made difficult by the historical context and turbulent environment outlined above.

It is assumed that such a continuum, by definition, is multidimensional since individual needs (based on a series of characteristics) must be matched to the proper service equivalent. Thus, for example, the ideal placement maximizes or minimizes certain patient characteristics in a service setting that is cost effective.

As a working approach, it is hypothesized that system rationalization for the provision of care for the chronically impaired elderly can be facilitated by developing a continuum of three dimensions. First, four sets of individual characteristics are identified that affect singly or, in combination, the older person's functional capacity. The four include: physical and mental health status, financial resources, informal supports, and physical environment.

Second is the dimension of resource absorption required to provide the living arrangement and service package prescriptive to the individual's functional capabilities. Third, is the dimension of

personnel requirements to staff the living situation to provide the needed services.

These dimensions are ordered along a continuum from high level of functional disabilities, high resource absorption in the provision of service, and high staff and technology requirements to the low end of the continuum in which the older person is able to maintain independent living and thus requires a minimum allocation of institutional resources and service provisions.

Some examples may illustrate the approach suggested here. First, there are the characteristics of older people that are critical determinants of their needs. At the low end of the continuum, are the older people who live, characteristically, with spouse or alone, in their own home. They may indeed have health problems (arthritis, diabetes, arteriosclerotic heart disease (ASHD) for instance); but, for the most part, these conditions do not substantially limit their abilities to perform the routine tasks of every day living.

They use private physicians, dentists, pharmacy services and may, at times, require acute treatment hospital stays of limited duration. Characteristically, a portion of their income comes from the Social Security system, and some of their health care expenses are covered by Medicare. They take advantage of reduced fare and other age-related income transfer and service entitlements. The majority of these people are still part of family and friendship groups, and the system of exchange between them and the other members of these groups is one of mutuality.

These people may choose to move from the home of their middle years to a smaller apartment or to a retirement community. The move in such cases is an "elective" having to do with items like reduced space needs, changing life style, sociability, etc. It is never, however, of the same order as a move to enriched housing or a senior citizens hotel or other type of domiciliary care facility which are examples of service-rich and more protected environments.

At the opposite end of the continuum are the chronically disabled: mental impairment may have become so severe that constant supervision and personal assistance is required. Or, a combination of ASHD, emphysema, and diabetes mellitus may have rendered the older person dependent on daily personal assistance and close medical and nursing supervision.

The health problems that are associated with a high degree of functional incapacity are, as we have noted, more likely to occur among the oldest of the 65+ cohort—those 75 and older. This is also the

group in which the incidence of poverty is greatest. Typically, they have been retired for a decade or more. Their Social Security benefits are lower than the benefits of the currently retired. If they have private pensions, it is unlikely that their pension benefits have been adjusted to take into account the erosion of purchasing power caused by inflation. Additionally, savings are depleted over the years to supplement monthly income or to meet medical care costs that are not covered by Medicare.

Thus, impaired functional capacity is often associated with declining and limited financial resources and, at the same time, there may also be erosion of the informal supports of the older person. Widowhood is the fate of the majority of women in the United States who live into their eight and ninth decades and most of these women live alone. Noted before were certain demographic trends which are adversely affecting the capacity of family members to provide daily hands-on-care to their functionally disabled relations. Support from friends and neighbors may similarly decline over the later years in the life cycle. Friends of the same age become ill, move away, die. Neighborhoods decline, their populations change, and the neighbors on whom the aged person relied upon may move away. The very old widow living alone and without children or other relatives nearby is frequently the most vulnerable among the aged population, and the most dependent on formal organizations and professionals for the services needed.

The second dimension of the continuum of care provides a picture of the living arrangements and service package prescriptive to the varying degree of functional incapacity. Ideally, certain conditions should be met in making the prescription:

First, the older person should receive the care and services needed in the least restrictive environment possible.

Second, decisions should be made on the basis of a careful multidimensional assessment of health status, social and psychological resources, etc., and the older person and family should participate in the decision.

Third, there should be provision for periodic and regular re-assessments so that changes in the health status and situation of the older person can be reflected in appropriate changes in the service package.

Fourth, access into the service network and movement within it must be available to all older people.

Already indicated are some of the obstacles—the fragmentation and discontinuities in service and benefits, the complexity of the system that makes it inaccessible to many, the variations and inadequacies in many assessment procedures—that block the path of functionally disabled older people to the services they need.

The consequences of our inability to provide rational, accessible, and comprehensive long-term care are apparent at both the macro and micro levels. The 1979 New York State Health Plan Final Report includes a section on long-term care which catalogues the consequences at the state level. “The long-term care system’s high cost precludes expansion in its current form,” the report concludes. This conclusion is a clear signal that future funding of services will depend on changes in it that will guarantee access and at the appropriate service level.

Paradoxically, the acute care hospital represents a critical juncture in long-term care systems’ rationalization. While these facilities offer the professional expertise and sophisticated technology to address the most complex diagnostic and therapeutic problems of the chronically impaired elderly, they often contribute to the fragmentation and discontinuity noted above.

Acute care hospitals are frequently the entry point into the long-term care system. Yet they have failed to implement mechanisms that emphasize and foster continuity of care between pre-admission, acute care, and posthospitalization services. Such deficiencies are exemplified by lack of ongoing involvement in the social-health network used by the elderly, and by the limited development of transfer agreements with long-term care facilities beyond the often superficial requirements of regulatory agencies. Predictably, therefore, the outcome of an episode of acute care hospitalization for an elderly individual is often a disposition crisis solved by transferring the individual to an expedient rather than appropriate setting.

In this context, it is the responsibility of the medical school to include in its long-term care agenda, an obligation to facilitate continuity of care in its affiliated hospitals via program development efforts. Such efforts might focus on, among other areas: development of medical information systems across involved facilities and providers, model projects to promote interprofessional intake and timely discharge planning with the objective of identifying the most appropriate site for posthospital referral; methods for monitoring quality of care during an episode of illness within which the patient might spend time in several types of facilities, including

CONTINUUM OF CARE

Functional Capacities <i>Physical and Mental Health Status</i>	<i>High Dependency</i>	<i>Low Dependency</i>
	Chronic and seriously disabling physical and/or mental health problems, requiring regular and frequent medical care; daily personal assistance and regular skilled nursing supervision. (Ex. Mental impairment—moderate to severe; seriously impaired eyesight; A.S.H.D. of sufficient severity to limit ability for self care)	Social and mental health status requires a relatively protected or service rich environment, so that older person receives assistance with specified tasks; and/or lives or spends significant amounts of time in a structured social environment.
<i>Financial Resources</i>	Living expenses and costs of medical and social services exceed fixed income and assets of older person (including Social Security and Medicare), old person eligible for publicly funded income tested programs (ie, S.S.F., Medicaid, etc.)	Older persons income and assets (in form of wages, Social Security and private pension benefits, savings, etc.) is adequate. Health care costs are partially covered by Medicare, and older person uses other age-related benefits (ie, half fare public transportation, tax benefits, etc.)
<i>Informal Supports</i>	Informal supports from family, neighbors and friends are functionally or absolutely absent or if present require substantial help from formal organizations over a sustained duration of time.	There is continuity in older person's relationships with family, friends, and neighbors; systems of reciprocal exchange among them; and although there are losses due to relocation, failing health, or death, the integrity of these groups is not seriously threatened.
<i>Physical Environment</i>	Living environment requires special adaptation to compensate for older person's disabilities, (ie, one floor housing unit with easy access to outside; wide doorways to accommodate wheelchair; hospital bed and other special furniture and equipment; handrails, etc.)	No significant environmental modification is required.

Resource Absorption	Skilled Nursing Facilities	Health Related Facility	Day Hospital Programs,	Enriched Housing	Senior Citizens Apartments	Independent Living in Community
	In the community with daily, round-the-clock home health services under skilled nursing supervision; acute treatment hospital staff and frequent use of physicians services; special furniture, equipment, etc. (ie, hospital bed; wheel chair, handrail, etc.)	Some use of acute treatment hospitals and/or SNF, reimbursement from combination of public entitlements and own income assets.	Telephone Reassurance Information Referral Advocacy Meals on Wheels as needed for health and home care service on a regular but limited basis. Age related and some income—tested benefits.	Telephone Reassurance Meals on Wheels Senior Center Nutrition Programs Information and Referral Advocacy Occasional Home Care Services and Home Health Services as needed Age related and income tested benefits (half-fare, etc.) Use of private physician, etc. and acute treatment hospitalization as needed.	Telephone Reassurance Senior Center Nutrition Programs Information and Referral Advocacy Occasional Home Care and Home Health Services as needed Age related benefits (half-fare, etc.) Private physician and acute hospital stays as needed.	Senior Centers Use of Private physician acute treatment and other health services as needed Age-related; but no income tested benefits
Staff Requirements	High staff/service user ratio, high degree of specialization and technology, physicians full range of specialties Nursing Staff (R.N., Nurse Practitioners, LPN's Aides) Social Workers, full range of other auxiliary services—x-ray, laboratory, pharmacy, P.T., O.T. recreation, dieticians, etc. Large support staff	Medium staff service user ratio, some specialization; modest technology, modest requirements for medical and nursing staff and medically oriented auxiliary staff. High requirements for social services, P.T., O.T. recreation and activities staff Medium support staff	Medium staff service user ratio, some specialization; modest technology, modest requirements for medical and nursing staff and medically oriented auxiliary staff. High requirements for social services, P.T., O.T. recreation and activities staff Medium support staff	Medium staff service user ratio, some specialization; modest technology, modest requirements for medical and nursing staff and medically oriented auxiliary staff. High requirements for social services, P.T., O.T. recreation and activities staff Medium support staff	Low staff/service user ratio, specialization but not at high level; social services, education, recreation, etc., specialists. Low support staff requirements	

feedback to the multiplicity of providers involved; and prospective studies comparing outcomes from alternative long term care treatment strategies.

*Mount Sinai School of Medicine/Hunter College
Long-Term Care Gerontology Center*

The need to engage academic institutions in the process of rationalizing the long-term care system has been recognized at both federal and state levels of government and, in the spring of 1979, the Administration on Aging of the Department of Health, Education, and Welfare issued guidelines for the establishment of university-based Long-Term Care Gerontology Centers.

The Administration on Aging described these centers as:

"a lead element of a new, comprehensive health and social service delivery system that is uniquely responsible to the multiple, fluctuating, and interacting needs and disabilities of the chronically impaired elderly."

AoA's intent is clear: the Centers are to be university based; a central component is to be the school of medicine; and there are to be functional and contractual arrangements between the academic institutions and service agencies.

Mount Sinai School of Medicine and Hunter College are both part of The City University of New York and have a long history of collaborative work, particularly in the education of social work students for work in the health care system. The experience of the two institutions, therefore, provide a solid foundation on which could be built a Long-Term Care Gerontology Center. In October 1979, Mount Sinai and Hunter were awarded one of 22 planning grants made by the Administration on Aging. There are wide variations among the 22 institutions, with respect to location of the centers within the university and the nature of the arrangement with service agencies.

In the planning process that preceded submission of the Mount Sinai-Hunter proposal, it was clear that an area of considerable strength for both institutions was the close working relationship between them and agencies and institutions in the service network. Mount Sinai and the Jewish Home and Hospital for Aged had just formalized an intent to an affiliate. In addition, Beth Israel Medical Center, Bronx Veterans Administration Medical Center, City Hospital Center at Elmhurst, Hospital for Joint Diseases Orthopedic Institute, and Joint Diseases North General Hospital all are teaching hospital affiliates of Mount Sinai School of Medicine.

Hunter College, particularly through its Schools of Nursing and Social Work and the Brookdale Center on Aging, similarly has working arrangements with a wide range of service agencies. The decision was made, therefore, to establish a Center structure and planning process in which service agencies representing all of the components of the continuum of care would have a central role. The Visiting Nurse Service, the major home health care agency in New York City, and the Jewish Home and Hospital for Aged were designated as Center affiliates, and a number of service agencies, in addition to the already listed medical centers, became Institutional and Research Centers. These include A. Phillip Randolph Senior Center, Casita Maria Incorporated Senior Citizens Program, Consumer Action of Bedford-Stuyvesant which operates both a long-term care facility and a home health care program, Community Service Society of New York, Friends and Relatives of Institutionalized Aged, Greater Harlem Nursing Home, Jewish Association for Services for the Aged, Marcus Garvey Nursing Home, and Search and Care. The participation of these agencies and institutions guarantees representation not only of the continuum of care; they also reflect the ethnic diversity of the City and ensure that ethnic differences which affect service delivery and utilization will be taken into account.

The planning task has been divided into six discrete, albeit interconnected work areas: professional education, research and evaluation, patient assessment, model projects, technical assistance, and public information. Committees were appointed to work in each of these areas and are composed of faculty from the School of Medicine and The College and staff from the affiliates and Instructional and Research Centers.

The agenda for these committees is to identify approaches and strategies that may yield innovations in the long-term care systems and promote diffusion of innovations among the providers, policy makers, and consumers and, finally, that will enhance the curricula of the professional schools which educate their students for the provision of care to functionally disabled older people.

Additionally, it is important to note the development of the Division of Geriatrics in the Department of Medicine at Mount Sinai. The objective of the Division, working through The Mount Sinai Hospital and the Jewish Home and Hospital for Aged, is to undertake the lead on behalf of the School of Medicine in addressing the fragmentation and discontinuity noted above as it applies to the acute care hospital setting. Through curriculum development, reorganization of clinical services, and

clinical and health services research, a concerted effort will be mounted to direct available resources toward promoting rationalization of long-term care services.

For example, an Acute Geriatrics Evaluation and Treatment Unit will be established not only to teach medical, nursing and social work students, but to demonstrate a coordinated, progressive, evaluative and therapeutic approach to selected elderly patients who, on admission, are at high risk for long-term institutionalization. Patients with potentially reversible disabilities and illnesses will be

admitted and treated, and a coordinated plan will be developed for community or long-term care facility services as required. Similarly, and with parallel and integrated development, the Division of Liaison Psychiatry will be developing a program to teach health workers how to deal with patient and staff reaction to geriatric care, and focus on knowledge, skill and attitude building concerning the geriatric patient in the clinical setting.

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Research in Geriatric Medicine at the Jewish Home and Hospital for Aged, New York—1944-1980

MANUEL RODSTEIN, M.D.

Introduction

Over the course of the past 110 years since its founding in 1870, there have been significant contributions to medical knowledge concerning the aged by the staff of The Jewish Home and Hospital for Aged as exemplified by many of the articles in this special issue on geriatric medicine of The Mount Sinai Journal of Medicine. We anticipate that with the affiliation of the Home and Mount Sinai School of Medicine there will be important collaborative publications enriching our knowledge of geriatric medicine and leading to further advances in the care of the aged.

Since 1942, The Jewish Home and Hospital for Aged has had an organized medical staff with diagnostic, radiology, and laboratory facilities and has accumulated a series of close to 3,000 necropsy examinations. Almost all of the attending staff then, headed by Frederic D. Zeman, held independent Mount Sinai Medical Center appointments as do a majority of the present attending staff.

The Home represents the largest nonprofit long-term facility for care of the aged in the United States. It offers a vertically integrated system of care for over 1800 aged residents at six levels of functional status. In ascending order of need, these are enriched housing in the community, a geriatric outreach program, a day care program, middle income housing, health related (intermediate) care, and skilled nursing care.

Residents ordinarily are followed for the rest of their lives after admission, 94% dying in the insti-

tution. Almost all of the remainder die at adjacent general hospitals, that is Mount Sinai and Montefiore, while under the care of attending physicians on the staff of the Home. This offers opportunities for the study of the natural history of disease with clinical pathologic correlations, emotional reactions to stress, and the evolution of the mental and physical aspects of the aging process.

Research is facilitated by a system of comprehensive initial and annual evaluations of each resident embracing all disciplines involved in their care.

I will now detail briefly some of the over 280 publications originating from the medical department of the Home over the past 36 years.

The frequency of silent and asymptomatic pneumonia in the aged was described in a series of 166 cases in 1946 (1).

A 1974 study of 250 consecutive necropsies (representing a 70 percent rate) showed that bronchopneumonia was the most common undiagnosed cause of death (2). In two studies of the mortality associated with chronic brain syndrome, bronchopneumonia was found to be the most frequent cause of death among the aged (3, 4).

A study of cancer revealed that the onset was often insidious without typical symptoms. Commonly, there was the discovery of anemia, and weight loss on a routine examination with the cancer found on further workup in a resident with few or no symptoms (5).

The importance of determining an individual's functional capacity rather than merely enumerating disease diagnoses was first established by Frederic D. Zeman in 1947. He described a system for classification of functional and therapeutic capacities that anticipated the attention given this problem by investigators and regulatory authorities many years later (6).

From the Jewish Home and Hospital for Aged, New York.

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In 1963, Dr. Zeman and the author correlated the presence and degree of arcus senilis with various indices of atherosclerosis. They found a relationship primarily with age and male sex and secondarily with the degree of coronary and retinal atherosclerosis (7).

In 1955 and 1964, in papers dealing with common clinical errors in the care of the elderly, Zeman pointed out many of the unusual clinical characteristics of heart disease in the aged, novel at the time, but now accepted axioms (8, 9).

A 1956 report on elderly residents of a long-term facility who had had nonfatal acute myocardial infarctions revealed that silent and atypical infarctions predominated among those over 80 years of age (10). In this group there was either no pain or a substitution symptom of chest pain, such as shortness of breath due to left heart failure with pulmonary congestion, dizziness, faintness, and confusion caused by a diminished cerebral blood flow and midabdominal distress due to backward failure.

In a clinicopathologic study of cases of myocardial rupture secondary to acute myocardial infarction, the incidence was unusually high among the elderly, particularly females, and especially prevalent among those in whom prior high blood pressure continued after the onset of an attack (11).

A double blind study of a group of residents who had been on long-term maintenance therapy with digoxin for unknown or transient indications revealed, over the course of 22 months of follow-up, that digoxin had to be reintroduced only because of an acute arrhythmia or the onset of acute failure secondary to acute myocardial infarction (12).

The life history of calcific aortic stenosis was studied in a clinical pathologic study of a prospective nature (13).

A psychological study of matched groups of aged with and without electronic cardiac pacemakers revealed remarkable long-term improvement in personality, adaptation and life style (14).

Orthostatic hypotension, a condition often accentuated by psychopharmacologic and antihypertensive drugs, was found to be common in the normal and of greater degree among those with diabetic neuritis, large varicose veins, pernicious anemia and those who have been at bed rest (15).

The tricyclic antidepressants even in usual dosages, have found to cause dangerous cardiac arrhythmias, conduction disturbances, acute coronary insufficiencies and myocardial infarction in the aged with organic heart disease (16).

In the field of ophthalmology, a basic reference study in 1957 by Kornswieg et al described the

visual capacities of more than 1,000 aged individuals (17).

Other studies described techniques of ocular surgery, the diagnosis and medical treatment of glaucoma, the function of a low-vision clinic, the pathology of diseases of the macula in the aged, diabetic retinopathy, and the prevention of blindness in the aged (18).

Walter G. Klopfer, the first psychologist to work in a long-term care institution, in 1951 described the role of the psychologist in facilities for the aged (19).

The current interest in American attitudes toward death and dying was anticipated in 1958 by Shrut in a paper entitled "Attitudes Towards Old Age and Death" (20).

Among the first psychiatric services in a long-term care institution for the aged was that initiated by Alvin Goldfarb at The Jewish Home and Hospital for Aged, New York, in the early 1950's. His work included development of the mental status questionnaire and the application of the face-hand test for the diagnosis of chronic brain syndrome in the aged. Dr. Goldfarb and his staff were at the forefront in the introduction of brief psychotherapy as a tool in the care of old people (21). His understanding and description of the psychiatric problems of the aged has been of incalculable value in the training of staff personnel in institutional settings (22).

The nature of long-term care institutions allows the selection of groups of elderly persons matched for age, sex, and medical status, with or without chronic brain syndrome, for participation in studies of the effect of the chronic brain syndrome on mortality and morbidity. The results of such studies have indicated an adverse effect on mortality of chronic brain syndrome per se and also increased mortality from pneumonia (4, 5).

Comprehensive reports on the precipitating physical, emotional and environmental causes of accidents in the institutionalized aged have stressed the significance of the aging process and disease in the increased accident liability of the aged (23, 24).

The sum total of these and related studies have resulted in appreciable contributions to the knowledge of the aging process and disease. In many ways, research of this type is good care of the aged. We expect that through the affiliation with Mount Sinai Medical School and Medical Center that the benefits of program of care and research at The Jewish Home and Hospital for Aged will provide a favorable environment to help train a future generation of geriatricians.

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A Medical School Curriculum in Gerontology and Geriatric Medicine

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The need for medical students to learn about the care of the elderly can no longer be ignored. Through the incorporation of gerontology into the basic science curriculum and geriatric medicine into the clinical experience, this goal can be achieved. Gerontology is the scientific study of aging from the biologic, psychologic, sociologic and demographic viewpoints. Geriatric medicine is that branch of medicine concerned with the clinical, preventive, remedial, social, educational, and research aspects of illness in the elderly in the long-term as well as the acute phase. The settings for the teaching of geriatric medicine include the acute hospital, the home care program, the long-term care facility and the ambulatory care clinic.

The current and future demography, epidemiology and health care costs of the elderly population are compelling factors for the incorporation of gerontology and geriatric medicine into the medical school curriculum. Elderly people while accounting for only 11% of the population receive 25% of all prescriptions and an even higher percentage of non-prescribed drugs (1). Thirty percent of the average internist's office visits and 18% of the average family practitioner's office visits are by patients over the age of 65 (2, 3). Forty-five percent of patients hospitalized by internists are 65 years of age or older (4). At least thirty percent of all acute medical and surgical hospital beds are occupied by elderly people at any given time (5). Currently, 95% of the 1.2 million nursing home's beds are occupied by people over the age of 65 (6). Their median age is 82.5.

Medical costs for patients over 65 are approximately three times as high as for those under 65 and account for 30% of the total personal health care expenditures (7).

When the current class of first-year medical students has reached the age of 60 (the year 2020), there will be 43 million people over the age of 65, twice as many older people as there were the day the student entered medical school. At that time, his/her practice of internal medicine or primary care will consist of a large proportion of elderly over the age of 75, a group with increased needs for health care services. With the number of dependent elderly growing, the future physician will see a growth in the number of nursing home beds despite efforts to increase home care programs and community services (8). The number of elderly people occupying acute hospital beds and requiring ambulatory care services will also rise (9).

Not only are there pressing economic and health care reasons for the introduction of gerontologic and geriatric medical material into the curriculum, but there are compelling intellectual and research questions. Our clinical knowledge has left many unanswered questions about the aging process and diseases occurring in late life. We do not know, for example, the cause(s) of senile dementia of the Alzheimer's type (SDAT), that accounts for approximately 50% of all cases of late life dementia (10). Nor do we know a successful treatment. We have not arrived at a cost effective and safe method to reduce the incidence of hip fractures in the elderly white female, the population predominantly at risk from osteoporosis. We do not know the relationship between adequate nutrition and normal aging (11). We know relatively little about the role of the aged liver in the metabolism of drugs or of changes in drug absorption that occur with age (12). Why elderly people have an increased sensitivity to some drugs has not been adequately ex-

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plained. The role of the immunologic system in carcinogenesis and the effect of aging upon it need expansion (13). Each of the organ subspecialties has many significant research questions that need to be answered if the quality and quantity of life in old age is to improve (14). Based on his intellectual needs, the demographic and epidemiologic facts, and economic pressures, the physician of the near future must have the knowledge base, positive attitude and clinical skills to care for the increasing number of elderly.

Medical Ageism

While the need for education about the elderly is recognized, its actual implementation may be complicated by ageism, a prejudice against an elderly person merely because of his advanced years (15). Ageism, like sexism or racism, manifests itself by individual, institutional and societal biases and pressures. These include, for example, pressure for early retirement, the belief that older people are less productive than younger ones, that they are unyielding and fixed in their attitudes, universally lose their memory, and eventually develop dementia.

Health care professionals are not immune to ageism. Physicians can develop, during medical school and residency training, a variant of ageism, medical ageism (16), which manifests itself by an avoidance (emotional, intellectual, and physical) of elderly patients. A student may reject internal medicine as a career choice because of his/her dislike for working with elderly patients.

"Crock," "gommer" and "turkey" are terms that most medical students have heard by the end of their third year. These terms are used to refer to elderly patients with confusional syndromes, incontinence, strokes, dehydration and/or immobility, all common and often reversible illnesses. In such an environment, medical students can become pessimistic about the outcome of therapeutic intervention especially when diagnostic evaluations are not pursued because the patient is "too old." Unfortunately, medical ageism can lead to a denial of the facts: Eighty percent of all elderly stroke patients will return with partial or full function to their homes (17); up to 50% of all acute confusional states may be reversible if properly diagnosed and treated (18); congestive heart failure, so commonly seen in the elderly, may be iatrogenically induced by digoxin (19) which can often be discontinued on an ambulatory basis (20, 21); the most common and easily reversible cause of fecal incontinence is a

fecal impaction; and a not insignificant proportion of elderly patients with chronic organic brain syndrome may have reversible causes including drugs and/or depression (22).

Medical ageism also manifests itself by the attitude that old people cannot have diseases that are interesting or worthy of discussion in medical schools or university hospitals. Clearly, this is not so. A review of the outstanding textbooks in geriatric medicine will put to rest such ideas (23-26). In 1978, the *New England Journal of Medicine*, devoted 12 of its 45 adult CPC's (27%) to discussing the clinical course and pathologic correlations of patients 65 years or older. The high percentage is remarkable in light of the difficulty obtaining post-mortem examinations in the elderly. Teachers should capitalize on the medical, psychiatric and social problems of their elderly patients in all phases of health and illness, separate the interrelated and complex problems, seek out the reversible components, elaborate on the body of knowledge, pose solutions for both acute and long term care and point out the unanswered research questions.

An Integrated Curriculum

The curriculum in gerontology and geriatric medicine must be comprehensive, multidisciplinary, integrated and required of all medical students (27, 28). In addition, it must be taught as a normal part of the life cycle. It must provide all students with: 1) a core of knowledge about these special areas, 2) opportunities for developing skills in caring for the elderly regardless of the phase of illness (acute or chronic), 3) a positive attitudinal approach to caring for the elderly, and 4) an understanding of the unanswered research questions. This material should be presented as a free standing, multidisciplinary course that draws faculty from the entire medical school. An alternative approach would be to integrate the gerontologic and geriatric curriculum into already existing courses in the form of lectures, portions of lectures, bibliographies, audiovisual material and exposure to both the well and ill elderly in all phases of health and illness. Specific questions based on the gerontologic and geriatric material should be included in written and practical examinations.

Material about aging is currently being presented formally in the following courses during the first two years: physiology, pharmacology, endocrinology, the course on clinical evaluation of the patient (history and physical examination), community

medicine, pathology, and human behavior. These courses feature summary lectures on the relationship of aging and the particular subject. These lectures could be presented early in the course, since it will give the student a "geriatric" framework with which to incorporate all future material. The student must learn to ask himself about each topic, "How does aging influence it?" and about every disease, "Does it have different ramifications (presentation, diagnosis, therapy) in the elderly?" All lecturers should deal with the gerontologic or geriatric aspects of their particular topic.

An outline of an integrated curriculum in gerontology and geriatric medicine, addressing itself to six major areas, is presented in Table I.

Clinical Settings

It is important to expose first and second-year students to healthy, community residing elderly people so that their possible biases can be confronted; that is, not all elderly are sick, dependent, demented, hospitalized, or living in nursing homes. Currently, a first-year elective, entitled "Introduction to Aging," is being offered. It allows the student to evaluate his/her attitudes toward the elderly and attempts to explore and positively influence them by participating in supervised interviews with healthy, community residing elderly as well as acutely or chronically ill elderly, in the hospital, nursing home, or home care program. Selected readings and discussions cover demographic, epidemiologic, health and social issues.

Clinical geriatric material must be integrated into the third-year clerkship especially in medicine, psychiatry, surgery, community medicine, gynecology, neurology, orthopedics, urology and anesthesiology. In addition, each of the departments should identify a faculty member who will be a clinical role model responsible for developing, integrating and implementing didactic and bedside teaching.

During the fourth clinical year, a mandatory subinternship in geriatric medicine should be developed that involves the medical student in a supervised primary care experience in a comprehensive health care delivery system for the elderly. Currently, an elective subinternship in geriatric medicine is being offered for fourth-year students based, in part, at the Jewish Home & Hospital for the Aged.

Geriatric material should be presented in the following ways during the fourth-year subinternship:

- 1) case presentations emphasizing unusual as-

pects of history taking, physical examination, altered symptomatology, multiplicity of illness, differential diagnosis and therapeutic decision making;

- 2) geriatric and gerontologic seminar series covering topics in geriatric medicine, rehabilitation medicine, geropsychiatry, surgery, and the health care system;
- 3) audiovisual material (videotapes or audio slide presentations) that condenses into minutes the progress of a patient over months. This longitudinal technique lends itself well to showing the special approaches used in the rehabilitation process of the elderly patient recovering from a fractured hip, stroke, or amputation. In addition, it allows the student to see the positive outcomes of long-term rehabilitation efforts;
- 4) special teaching modules including educational objectives, pre and post tests, reading material and problem solving exercises that have been designed for use in various settings in which elderly people are cared for (Table II) (29).
- 5) involvement in team meetings and team process. The methodology and skills of the multidisciplinary team approach are taught as an integral part of the health care system for the elderly. The team may include nurse, social worker, physical and/or occupational therapist, pharmacist, dentist, podiatrist, speech pathologist, audiologist, physician, nurse practitioner, and physician assistant.

The subinternship, while primarily based at one type of health care institution (skilled nursing facility, acute hospital, or home care service), should expose the medical student to all phases of health and illness in both the community and institution (Table III). Emphasis is placed on learning the body of knowledge of geriatric medicine as well as the special approaches needed to deal with the complex problems of the ill elderly and their families.

Conclusion

The intellectual, demographic, epidemiologic, and economic reasons for the formal incorporation of gerontologic and geriatric material into the medical school curriculum are clear and pressing. Confronting the medical ageism of both students and faculty and positively affecting it, will be a challenging part of the process of introducing such a curriculum. That a fascinating body of knowledge

exists which needs expansion will then be discovered. An outline of an integrated curriculum in gerontology and geriatric medicine has been presented that can be the basis for independent courses

and/or be integrated into existing courses. We must prepare the medical student of today for both practice and research in the care of the elderly, the major health care consumer of the future.

TABLE I

Outline of Undergraduate Gerontology and Geriatric Medical Curriculum

- 1) Biology of Aging
 - a) theories of aging
 - b) mechanisms of aging
 - c) normal aging versus pathologic changes including anatomic, biochemical, physiologic, immunologic, neurologic, endocrinologic and sexual function changes
 - d) human growth and development (normal senescence)
- 2) Behavior and Neurosciences
 - a) psychologic changes (self perception, learning time, creativity, work performance, reaction time, memory, motivation, engagement versus disengagement)
 - b) life style and social changes (family support, retirement, losses versus disengagement)
 - c) depression, cognitive dysfunction, theories of cognitive decline and depression
- 3) Clinical Geriatrics
 - a) principles
 1. multiple pathology, underreporting, unusual presentation
 2. altered medical interviewing and history taking skills including mental status examination
 3. altered biochemical and physical parameters
 - b) pathophysiology, presentation and therapy of disease occurring almost exclusively in the elderly, for example, senile dementia, osteoporosis, accidental hypothermia, calcific mitral annular disease, polymyalgia rheumatica, hyperosmolar, hyperglycemia nonketotic coma, monoclonal gammopathy, and presbycusis.
 - c) pathophysiology, presentation and therapy of diseases or symptoms that occur in all ages but with special implications in the elderly, for example, hypertension, diabetes mellitus, stroke, pneumonia (prevention and therapy), depression, pulmonary emboli, weight loss, dizziness, urinary incontinence, asymptomatic bacteriuria, hypo and hyperthyroidism, silent myocardial infarction, and falls
 - d) pharmacology of aging including altered pharmacokinetics and pharmacodynamics, polypharmacy, drug-drug interactions and compliance
 - e) geriatric rehabilitation including therapeutic techniques and course of immobility, arthritis, fractured hip (operative and nonoperative approach), stroke, amputation and pressure sores.
 - f) surgery and anesthesiology of the elderly including peripheral vascular disease, urologic disease, colonic and breast surgery, plastic surgery, cholecystectomy, coronary artery bypass, and heart valve replacement
 - g) geropsychiatry
 1. acute (often reversible) organic brain syndromes
 2. chronic organic brain syndromes
 - a. senile dementia, Alzheimer's type
 - b. multi-infarct dementia
 - c. secondary dementia (occasionally reversible)
 3. affective disorders (depression, mania)
 4. psychoneurosis
 5. transient situational disorder
 6. paranoid state
 7. benign memory loss
 8. therapies (psychotherapy, family, group and drug)
- 4) Health Care System—especially designed for needs of dependent and high risk elderly
 - a) demography and epidemiology of the aged
 - b) preventive geriatrics—health care screening, preretirement screening, senior citizen health education, home visits to prevent accidents, nutrition and alcohol counseling; dental and podiatric screening
 - c) home care services—(5%–8% of all elderly are homebound)
 - d) geriatric component to ambulatory care clinic
 - e) acute hospital unit involving both evaluation and treatment
 - f) long-term care component (skilled nursing facility, health related facility)
 - g) support services
 1. informal (family, friends, neighbors)
 2. formal (community, agencies and services)
 - h) economics: Medicare, Medicaid
 - i) the multidisciplinary team approach
- 5) Ethical, legal and attitudinal issues
 - a) struggle to remain in community
 - b) informed consent and dementia
 - c) refusing care by patient
 - d) guardianship and conservatorship
 - e) denial of treatment option by physician
 - f) criteria for resuscitation
 - g) family pressures
 - h) options for health care—home or institution
- 6) Research Methodology
 - a) separation of age effects from disease
 - b) relationship of aging animal models to humans
 - c) longitudinal versus cross sectional studies
 - d) derivation of normal standards for elderly

TABLE II

Core of Didactic Material for Fourth-year Subinternship in Geriatric Medicine

Concepts of geriatric medicine
Interviewing and history taking in the elderly
Mental status examination
Medication and the elderly
Geropsychiatry
Rehabilitation of the elderly*
Hearing and the elderly
Thyroid disease in the elderly
Hypertension and the elderly
Speech and language disorders in the elderly
Geriatric dentistry*
Ophthalmology and the elderly
Social and economic aspects of geriatric care in the U.S.A.
Nutrition and the elderly
Podiatry and the elderly
* Audiovisual components available.

TABLE III

Components of Health Care System for the Elderly

Health care screening and education (senior citizen centers or preretirement settings)
Geriatric component to ambulatory care service
Acute geriatric evaluation and treatment unit in hospital or SNF
Skilled nursing facility (SNF) with active rehabilitation component
Home care service
Geriatric day hospital

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Age-related Weight Changes in Female C57BL/6J Mice: Nonpregnant Body Weight and Extra-fetoplacental Maternal Weight Throughout Pregnancy

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Abstract

A significant rise in body weight occurred between the ages of three and 11 months in female C57BL/6J mice given free access to food and kept under strictly controlled conditions in a limited access colony designed for aging studies. Thereafter, body weight remained stable until at least 21 months, the oldest age examined in this study.

Body weight was found to vary not only as a function of age, but also with past reproductive history. Previously pregnant mice weighed more than nulliparous animals, and this difference persisted at older age. The extra-fetoplacental maternal body weight (EFP-MW), (the maternal weight after removal of the uterus containing the entire conceptus) was likewise influenced by previous parity. The EFP-MW of 11-12 month-old mice was significantly higher in multiparous than in primiparous animals.

The pattern of increase in EFP-MW during pregnancy was similar in young (three-seven month-old) and in older (11-12 month-old) mothers. After relatively stable levels during the first trimester, EFP-MW increased by approximately 15% to a maximum at the middle of the third trimester. The subsequent decline was markedly delayed in the older mice, but both age groups reached their day 1 postparturitional levels at least 24 hours before delivery.

The regulation of EFP-MW by progesterone and possible neuroendocrine and peripheral sites of action of that hormone are discussed.

Introduction

Human body weight changes after attainment of maturity and reveals variable patterns depending on age, sex, diet, socioeconomic status, and other factors. In general, body weight increases during the middle years and declines at old age (1). A survey in the United States indicated that the rates of adult body weight changes differ between the sexes. Women showed weight increases over a considerably longer period of time than did men, and their subsequent body weight decline at old age was proportionally less (2). A Canadian (3) and a French (4) study found patterns of weight changes and differences between the sexes similar to those reported for the United States. A comparative study

between a group of Parisians and a North African Caucasian population found a middle age weight gain only in the Parisians, whereas the weight decline at old age was common to both groups (5), suggesting an effect of socioeconomic factors on human weight patterns.

Several studies in laboratory rodents indicate similarities in adult body weight changes with those observed in humans. White SAS/4 mice (6) and LAF₁ mice (7) showed a steady, low, age-related weight decline after a one-year maturational rise and a short plateau period. Similarly, noninbred Wistar rats exhibited a weight loss in the later period of their lifespan, amounting to 20% during the last 200 days before death (8). However, it has been pointed out that weight loss in laboratory rodent at old age may not be a true aging phenomenon, but a consequence of disease (9). This view is supported by a study that found weight decrements

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in male C57BL/6J mice only in a subgroup of animals with pathologic lesions, in contrast to healthy individuals that exhibited stable body weights beyond the age of 26 months (10). A subgroup characterized by extended longevity was also identified in a study of Sprague-Dawley rats. That subgroup retained stable body weights into old age, in contrast to a shorter-lived population characterized by heavier body weight and higher body fat content, which exhibited a marked weight loss during later life (11).

Considerations of body weight patterns throughout the human life span are of importance since there is evidence of an inverse correlation between weight and life expectancy (12), an observation that has been reinforced by results from experiments in laboratory rodents, based on early caloric deprivation to reduce growth rates (13, 14) and on qualitative variations in experimental diet (15).

Human maternal body weight during pregnancy rises gradually, beginning late during the first trimester (16), a rise that is only in part caused by the developing conceptus and the accumulation of fluids associated with fetal development (17, 18). In addition to water retention by tissues and an increase in blood volume, the extra-fetoplacental maternal weight gain is the result of augmented protein and fat deposition (19). The accumulation of protein becomes evident in the marked positive nitrogen balance, which was found to occur in excess of the needs of the fetus (20). In contrast, the period after birth is characterized by a negative nitrogen balance until return to the normal, nonpregnant state which, in humans, requires two to three months for its completion (20). The deposition of body fat appears to be highly variable among individuals, and losses as well as gains have been observed during normal human pregnancy in a study which concluded that the total body fat is regulated by the energy content of the diet, just as in the nonpregnant state; whereas the total body cell mass increases irrespective of dietary caloric values, provided an adequate protein supply is available (21).

Experimental animal models have provided valuable information about the hormonal control of the extra-fetoplacental maternal weight gain as well as the fetal role in the regulation of that weight gain. The extension of such data to humans, however, must be made judiciously since considerable inter-species differences exist (18, 22). Hormonal mechanisms of weight control have been most extensively studied in laboratory mice and rats, and progesterone has been implicated as the principal hormone involved in body weight regulation in

those species on the basis of the following observations. The pregnancy-related weight increment on day 15 or day 16 could be maintained by progesterone treatment after hysterectomy, with or without ovariectomy, or by injection of HCG or PMS, acting as luteotropic agents, if the ovary remained intact (22). When administered to nonpregnant mice, progesterone caused a weight gain comparable to that of pregnancy, and an extensive weight reduction was observed after cessation of treatment (22). A progesterone-induced weight gain was likewise reported for nonpregnant female rats; in contrast, the hormone was not effective in males (23).

The rapid postparturitional weight decrease in rodents is assumed to be related to progesterone withdrawal. In mice, weight maintenance does not appear to depend on the fetus, since destruction of mouse fetuses on days 12–15, with the placenta remaining in situ, did not diminish maternal body weight until the placenta were delivered at the normal time (24). Weight levels were likewise maintained in pregnant mice after adrenalectomy on days 13–15 (25), suggesting that the adrenal gland does not play a major role in the weight control during murine gestation.

The present study reports weight changes as a function of age and of previous reproductive history in female C57BL/6J mice. In addition, extra-fetoplacental maternal body weight patterns and their age-related changes are examined throughout pregnancy and during the puerperium. The regulation of extra-fetoplacental body weight is discussed in relation to previously reported plasma progesterone levels, that were surveyed throughout pregnancy (26) and during the preparturitional period (27) in the same animals as those of the present study.

Materials and Methods

Female C57BL/6J and male C3HeB/FeJ mice were obtained from the Jackson Laboratory, Bar Harbor, ME. On arrival, the females were either two-month-old virgins or eight-month-old retired breeders that is, animals that had been bred continuously for commercial purposes after attainment of reproductive maturity, and the males were two-month-old virgins. Before being used as proven breeders in the present pregnancy studies, they had fertilized at least one young female that subsequently gave birth to a healthy litter. The mice were quarantined for health screening and one animal of each new cohort was given a complete postmortem examination before addition to the main colony, which was a limited access colony

designed especially for chronic female reproductive aging studies, kept at a 12-hour light cycle and at $24 \pm 1^\circ\text{C}$. Details concerning the housing and mating protocols were previously reported (26, 28). Day 1 of pregnancy was the first day after exposure to a proven breeder male on which a vaginal plug was found during daily inspection before noon. Approximately 15% of the pregnant mice were eliminated because of gross pathologic lesions, some of which included 3% for blindness and gross eye defects that are characteristic of this strain (29), 4% for vaginal septa, 5% because of enlarged spleens. Whole body weights were measured to the nearest 0.1 gram either in conscious or in Avertin (1,1,1-tribromoethanol) anesthetized mice. The conceptus, that is, the uterus and its entire content, was weighed after removal under anesthesia, and the extra-fetoplacental body weight was determined by difference. The day 1 postpartum maternal weight, excluding the uterus, was obtained between 12 and 24 hours after parturition.

Statistical significance was determined by the 2-tailed Student t-test and by coefficient of correlation analysis.

Results

Table I presents body weights of nonpregnant female C57BL/6J mice of three different reproductive histories: virgins, young mature mice after one previous pregnancy, and retired breeders. In addition, the Table shows maternal body weights within 24 hours after parturition and weights of animals exhibiting a vaginal plug without subsequent pregnancy.

Body weights increased significantly in virgins between the ages of four and ten months (Group I vs II) as well as in previously pregnant mice during a similar lifespan, ages six and 11 months (Group III vs IV). No increase was found in 21-month-old animals when compared to 11-month-old mice (Group IV vs V).

TABLE I
Body Weights of Female C57BL/6J Mice as a Function of Age and Parity

Group	Age	Reproductive State	Type of Measurement	Body Weight grams $\pm$ SEM (no of mice)	Significance
I	4 month	virgins (Vi)	total body weight (TBW)	20.1 ± 0.3 (37)	
II	10 month	Vi	TBW	23.5 ± 0.4 (14)	I vs II: $t = 6.72$; $df = 49$; $P < 0.001$.
III	6 month	one previous pregnancy	TBW	22.9 ± 0.4 (21)	I vs III: $t = 5.99$; $df = 56$; $P < 0.001$.
IV	11 month	retired breeders (RB)	TBW	28.7 ± 0.3 (28)	III vs IV: $t = 12.28$; $df = 47$; $P < 0.001$ II vs IV: $t = 10.38$; $df = 40$; $P < 0.001$
V	21 month	RB	TBW	29.6 ± 0.7 (20)	IV vs V: N.S.
VI	11-12 month	first pregnancy	extra-fetoplacental maternal body weight (EFP-MW) d 8 of pregnancy	27.3 ± 0.8 (8)	
VII	11-12 month	RB	EFP-MW d 8 of pregnancy	$29.3 \pm 0.4^*$ (30)	VI vs VII: $t = 2.46$; $df = 36$; $P < 0.02$
VIII	3-7 month	second pregnancy ≤ 24 h postpartum	extra-uterine body weight (EU-BW)	28.8 ± 0.4 (18)	
IX	11-12 month	RB ≤ 24 h postpartum	EU-BW	32.6 ± 0.5 (22)	VIII vs IX: $t = 6.17$; $df = 38$; $P < 0.001$
X	3-7 month	second pregnancy† plugged-nonpregnant	EU-BW	24.4 ± 0.4 (21)	
XI	11-12 month	RB† plugged-nonpregnant	EU-BW	27.4 ± 0.3 (41)	X vs XI: $t = 5.57$; $df = 60$; $P < 0.001$
XII	11-12 month	first mating† plugged-nonpregnant	EU-BW	23.8 ± 1.0 (5)	XI vs XII: $t = 3.77$; $df = 44$; $P < 0.001$

* This value appears in Fig. 1 and is given here for comparison.

† These values represent the mean body weights, after removal of the uterus, of all 3-7 month-old mice between day 6 and day 19, and of all 11-12 month-old mice between day 6 and day 22 after discovery of the vaginal plug. Prior to day 5, implantation success could not be reliably determined by inspection of the uterus at laparotomy.

Nonpregnant weights in mice of comparable ages varied as a function of previous parity. Young, previously pregnant mice (Group III) weighed significantly more than virgins of a comparable age (Group I), and 11-month-old retired breeders (Group IV) were heavier than ten-month-old virgins (Group II). A difference in body weight as a function of past reproductive history was likewise observed in aged (11–12 months) pregnant mice: the extra-fetoplacental maternal weight on day 8 of pregnancy was significantly lower in mice kept virgins throughout the major part of their reproductive lifespan (Group VI) than in retired breeders (Group VII). A similar relationship was apparent between 11–12 month-old plugged-nonpregnant females after first mating (Group XII) and a group of retired breeders (Group XI).

Body weights of previously parous plugged-nonpregnant mice were significantly higher in 11–12

month-old (Group XI) than in three–seven-month-old (Group X) animals. Weights determined within 24 hours after parturition were also significantly greater in aged (Group IX) than in young (Group VIII) mice.

The extra-fetoplacental maternal body weights, presented in Figure 1, were significantly higher in 11–12 month-old than in three–seven month-old mice. This difference was observed on all days throughout pregnancy and amounted, on the average, to 15%. In the course of pregnancy, both age groups exhibited a gradual rise in extra-fetoplacental body weight from a mean weight of 24.6 g during the first six days to a maximum weight of 30.1 g (+22%) in young, and from mean 28.8 g to maximum 33.3 g (+16%) in older animals. The rise in both age groups started approximately at the beginning of the second trimester, and it appeared to take place more smoothly and somewhat earlier in the older

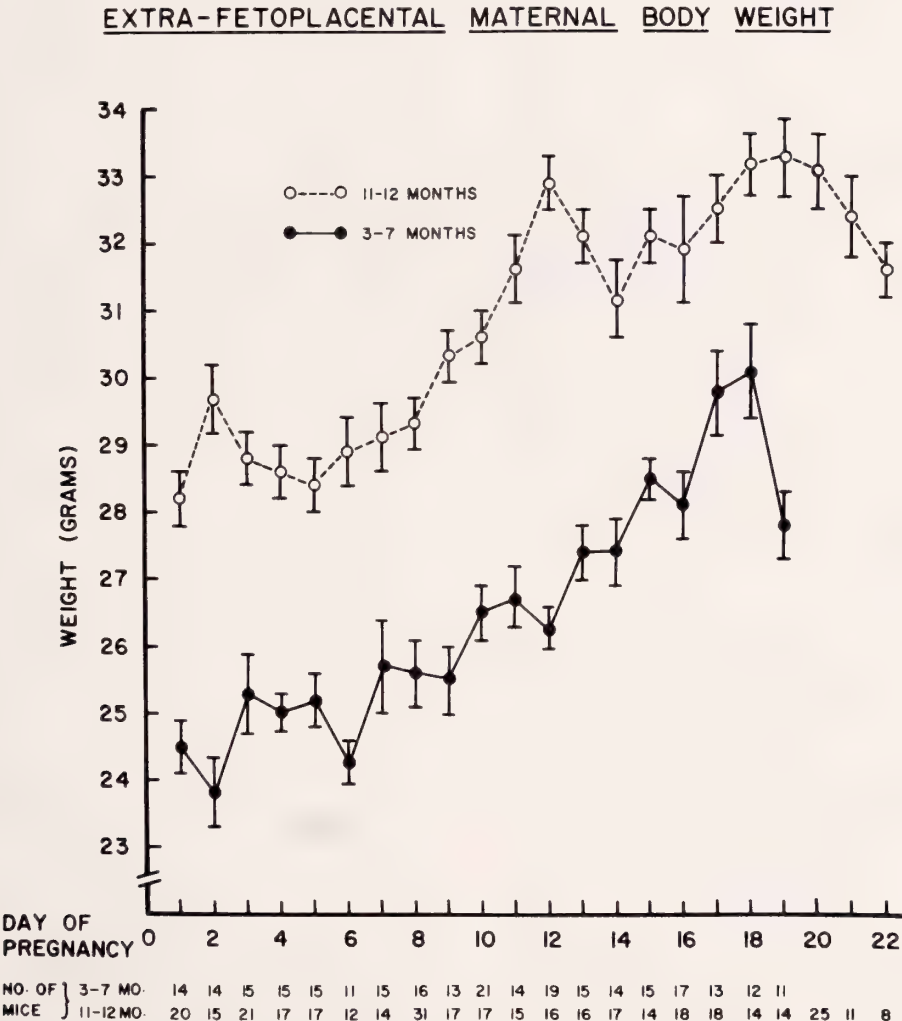


FIG. 1. Extra-fetoplacental maternal body weights in 3–7 month-old and 11–12 month-old pregnant C57BL/6J mice. (These weights were determined after removal of the uterus containing the entire conceptus).

mice. No marked age difference in the rates of weight gain were observed.

The preparturitional body weight decline occurred swiftly in young mothers, with a decrement of approximately 8% between day 18 and day 19. In contrast, the decline in older mothers took place gradually, starting on day 19. In both age groups, no further decline was observed within the first 24 hours after parturition (Table I, Groups VIII and IX).

The coefficients of correlation between extra-fetoplacental maternal body weight and previously reported progesterone levels throughout pregnancy (26) were calculated for all days of pregnancy. There was a significant positive correlation between rising progesterone levels and increasing body weights on day 12 in young mice ($P < 0.02$) and on days ten to 13 in aged animals ($P < 0.05$).

Discussion

The present data show an increase in body weight between the ages of three and 12 months in female C57BL/6J mice under conditions of freely available food. The observed rise in body weight, however, does not appear to be a simple function of age. Young mice after one pregnancy weighed significantly more than virgins of comparable age. This weight difference, as a function of past reproductive history, persisted during later life in mice of several reproductive states: ten-month-old virgins weighed less than 11-month-old retired breeders, as did 11-12 month-old mice on day 8 of their first pregnancy when compared to 11-12 month-old, day 8 pregnant animals, after repeated previous pregnancies (retired breeders), and 11-12 month-old multiparous plugged-nonpregnant animals were heavier than nulliparous plugged-nonpregnant mice of the same age.

The question arises whether the higher body weights in young mice after one pregnancy (compared to that of virgins of equivalent age) were caused by a slow return of their body weight to the nonpregnant level, or by a permanent alteration. The former possibility is unlikely since weight measurements were made no earlier than five weeks after delivery. Furthermore, 11-12 month-old retired breeders continued to show elevated body weights for over three months after their last delivery. It is, therefore, concluded that the hormonally regulated metabolic changes that occur during pregnancy result in a permanent increase in the subsequent nonpregnant body weight. This increase is retained until older age, that is, in the present

animals until at least 12 months and is, at that age, also observable in the extra-fetoplacental maternal body weight on day 8 of pregnancy. Whether the net increase in nonpregnant body weight as a result of previous parity is a function of the number of preceding pregnancies or a discreet event determined by the metabolic changes during the first pregnancy cannot be determined from the present data.

Our results do not support other observations (6-8) of a body weight decline in rodents during later life. This is likely to be due to the overall good health of the mice included in this study, as determined by the absence of pathologic lesions observed under the dissection microscope at necropsy. The importance of distinguishing true age-related physiologic changes from pathologic alterations, and the frequent failure of past gerontologic research to do so, has recently been emphasized (30, 31). Specifically, with regard to body weight changes in the present strain, the majority of aged healthy males, in contrast to diseased animals, was able to retain stable body weights at least up to 26 months of age (10), an observation similar to that reported for rats of both sexes (32).

In addition to pathologic changes, a number of environmental factors are likely to influence age-related changes, including those of weight, in animals of the same strain kept at different colonies. For example, longevity of C57BL/6J mice differed considerably in two separate colonies. Females and males showed a mean life span, respectively, of 692 days and 676 days (33) and of 794 days and 879 days (34).

Since body weights of laboratory rodents under several experimental conditions appeared to be increased by progesterone (22-24), it was of special interest in the present study to correlate extra-fetoplacental body weight patterns with previously reported progesterone levels throughout pregnancy (26) and at parturition (27). The initial threefold increase in plasma progesterone levels found in both age groups from day 1 (the day of the vaginal plug) to day 6 of pregnancy was not accompanied by a major rise in extra-fetoplacental body weight. However, the weight increase at the beginning of the second trimester appeared to reflect the second rise in circulating progesterone observed at that time. This was evident in the statistically significant positive correlation between rising progesterone levels and increasing body weights on day 10 to day 13 in the older, and on day 12 in the younger animals. Similarly, ovariectomized four-month-old and ten month-old mice that were injected with a sequence

of progesterone and estradiol, which mimics hormonal levels during the first eight days of pregnancy (35), did not respond with weight increases (Holinka and Finch, unpublished observations).

The preparturitional weight loss observed in both age groups approximated the pattern of preparturitional progesterone decline, which was considerably delayed in the 11–12 month-old animals (27). It is noteworthy, however, that in both age groups the weight decline followed the progesterone decrease by one to two days. These observations suggest that, if progesterone plays a role in body weight regulation during pregnancy, as suggested by the statistically significant correlation of the two parameters at midpregnancy and the similarity in their patterns of change thereafter, then the action of progesterone occurs in a delayed fashion. This is indicated both by the rises in body weight in response to earlier increases in progesterone levels, and by delayed weight decreases in response to progesterone withdrawal. Characteristically, in both age groups the postparturitional extra-fetoplacental body weight levels are reached at least 24 hours before parturition.

In conclusion, the patterns of extra-fetoplacental body weight increase, as a function of progressing pregnancy, are essentially unimpaired in aged mice, and the larger initial weights of the older animals reflect their higher nonpregnant body weights. Whether the delayed preparturitional weight decrease is a primary age-related phenomenon or, as is more likely, whether it reflects the delay in progesterone decline (27) and possibly in estrogen increase (36) observed in these animals remains open to further experimentation.

It may be speculated that the weight inducing effect of progesterone observed in female but not seen in male rats (23) was caused by a modification of hypothalamic-pituitary-thyroid mechanisms that resulted in alterations of basal metabolism. Such a possibility presupposes differences between the sexes in brain and/or pituitary progesterone target areas, as defined by the existence of high affinity, low capacity progesterone binding macromolecules (receptors). Experimental proof for the existence of progesterone receptors in the brain and pituitary, and of possible qualitative and quantitative differences in receptors between the sexes is, as yet, incomplete (37). A major modification of the brain-pituitary axis by progesterone during pregnancy seems unlikely, since mice hypophysectomized on day 10 of pregnancy increased their body weight normally (38).

Alternatively, it is possible that progesterone reg-

ulates the hypothalamic feeding centers and thus, feeding behavior, without a direct modification of the neuroendocrine axis. It is also possible that the hormone affects areas outside the classical target tissues, and that it does so to a different extent in the two sexes. In addition, the interaction of other hormones in the control of body weight must be considered. The task of assigning a definitive role to progesterone in the regulation of extra-fetoplacental maternal body weight becomes further complicated by the possibility that the effects of that hormone observed after exogenous injection under experimental conditions may differ considerably from its endogenous action during pregnancy.

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Metabolic Interrelationships During Aging: The Heme Metabolic Pathways*

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Abstract

The biochemical processes of aging are manifested at the molecular level by changes in enzyme activity. When such changes occur in key regulatory enzymes, the flux through a metabolic pathway is altered. As substrate and product levels in the affected pathway change, metabolic interactions, such as feedback inhibition, may cause these changes to be reflected in the activities of neighboring pathways. Thus, subtle and important disruptions in cellular metabolism can be brought about through long-term alterations in the activities of a few key enzymes. The present report describes our studies of metabolic interrelationships in the heme biosynthetic pathway during aging.

Introduction

During the past several years, much attention has been directed toward an understanding of the biochemical mechanisms and consequences of aging. A great impetus toward this goal has been the realization, chiefly through the work of Orgel (1), that age-related changes at key metabolic foci within the cell may initiate a chain of events that leads to biological collapse. When the "error catastrophe" hypothesis was originally proposed, the key event was postulated to be production of an altered enzyme whose impaired function would compromise the fidelity of protein synthesis initiating a degenerative cascade. While it is clear that the accumulation of many classes of proteins with altered amino acid sequences would seriously compromise cellular function, less dramatic changes in protein function might prove equally destructive.

As currently defined, aging represents a loss of the ability to adapt to the environment (2). At the molecular level, this adaptation involves changes in the activities of inducible enzymes. The degenerative alterations seen during aging may result not only from changes in the activities of isolated en-

zymes, but may more closely reflect a cascade of alterations in the flux through metabolic pathways. This could be brought about if an age-related change occurred in a single key regulatory enzyme. Under such circumstances, changes in the rate of flux through one metabolic pathway could affect the levels of intermediates and end products in pathways, that both precede and follow the affected pathway. Figure 1 illustrates this point. A decrease in the activity of the key regulatory enzyme not only decreases the availability of intermediates and end products from pathway 3, but it can change the rate of flux in numerous interdependent pathways (for example, feedback loop for pathway 1). Thus in theory, long-term changes in the activity of a single key regulatory enzyme could ramify in quite unexpected and important ways.

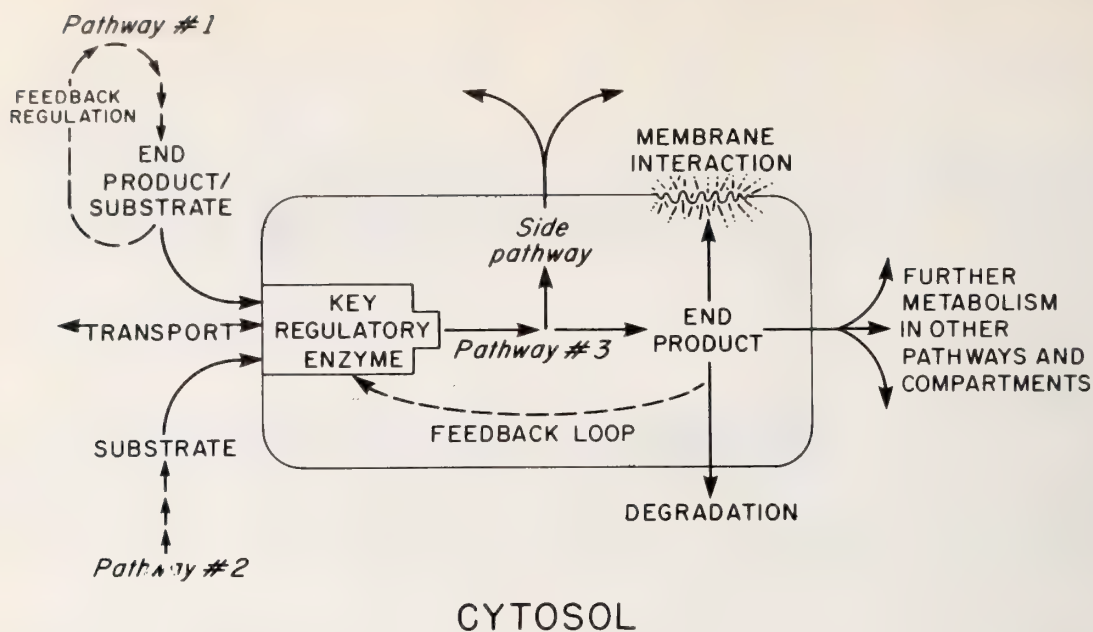
As an approach to elucidating the complex nature of age-related alterations in regulatory enzymes, we have begun to examine the regulation of heme metabolism during aging. The heme metabolic pathway contains several elements that can be exploited in studies of the metabolic consequences of aging (Fig. 2). These include:

1. A rate-limiting enzyme whose mechanisms of regulation are a subject of active investigation.
2. Cellular compartmentation of the overall biosynthetic pathway, with multiple membrane interactions of both intermediates and end-products.
3. An end-product, heme, which is at once, an important regulatory molecule, a prosthetic

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CYTOSOL

FIG. 1. A hypothetical scheme for the interactions between metabolic pathways involving a single key regulatory enzyme. Please compare with Fig. 2.

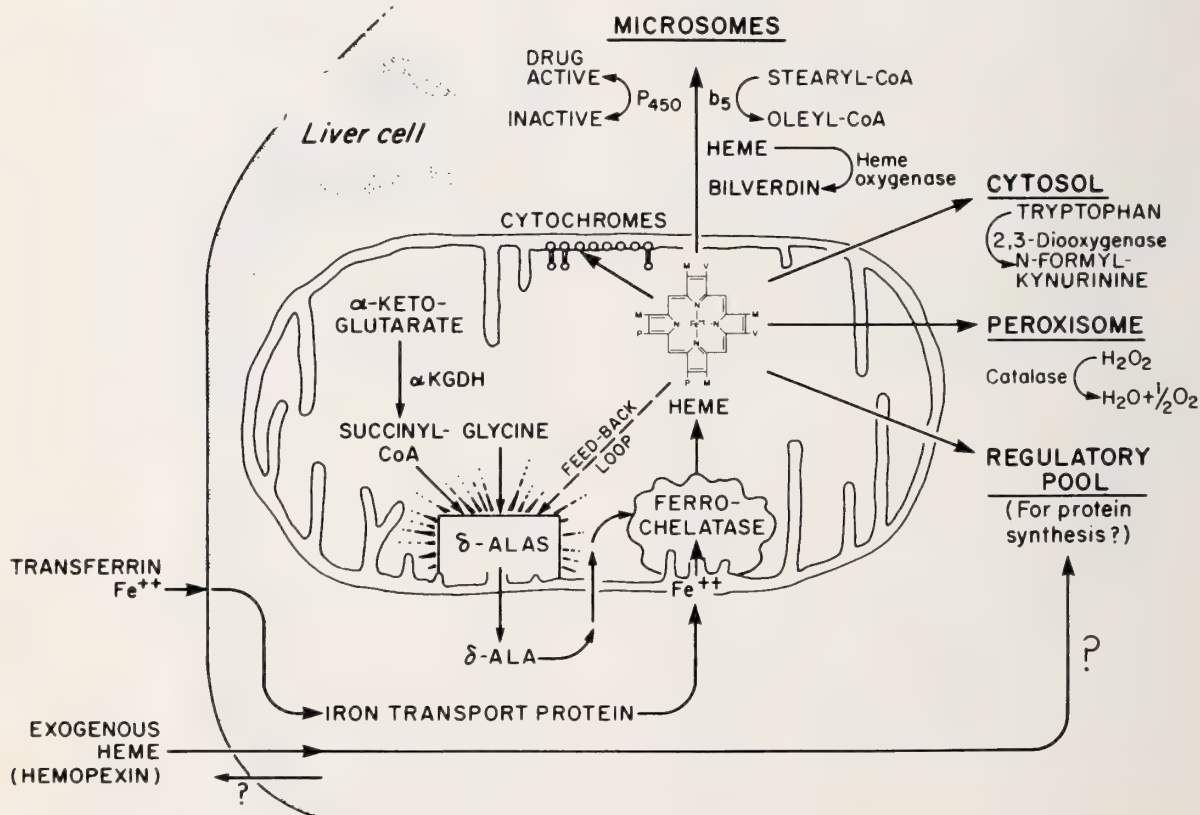


FIG. 2. The heme biosynthetic and metabolic pathways in the liver cell. Please compare with Fig. 1.

group for numerous apoproteins that are indispensable for cellular integrity, and a substrate for a specific degradative pathway. It will become clear that progress in understand-

ing certain of the above interactions has raised new questions that reflect the enormous scope and subtlety of the biochemical processes of aging (for example, the role of exogenous heme in liver heme

metabolism). We present this report as a map of the roads we have taken.

δ -Aminolevulinic Acid Synthetase

The overall rate of mammalian heme biosynthesis is regulated by the activity of the first enzyme of the pathway, δ -aminolevulinic acid synthetase (ALA-S) (3). This membrane-bound mitochondrial enzyme catalyzes the condensation of succinyl-CoA, synthesized in the mitochondria by the action of the Krebs cycle enzyme α -ketoglutarate dehydrogenase, with glycine to form the aminoketone, δ -aminolevulinic acid (4). In the rodent, ALA-S activity has been identified in a variety of tissues including kidney, heart, adrenal, spleen, and brain (5-9). The rat liver enzyme is the best characterized, and has been studied in homogenates (10), and isolated mitochondria (11), as well as in a purified preparation from rat cytosol (12). Recently, we have reported the purification, to homogeneity, of hepatic mitochondrial ALA-S (13). The activity of the liver enzyme has been shown to be affected by a variety of factors including nutritional (6, 7, 14, 15) and hormonal status (16-19). The activity is subject to induction by porphyrinogenic drugs (3, 20-22) or ethanol (23, 24), as well as probable feedback inhibition by heme (13, 20, 25, 26).

Once ALA is formed in the mitochondrion, it enters the cytosol for participation in pyrrole and tetrapyrrole formation. Subsequent intermediates again enter the mitochondrion for final tailoring prior to insertion of Fe^{++} into the ring of protoporphyrin IX by ferrochelatase to form heme (Fig. 2). Once formed, heme has many metabolic fates, in numerous cellular compartments. The cellular levels and distribution of heme should reflect the flux through each of these pathways. Thus, a number of workers (27-30) have postulated that the rate of hepatic ALA-S may be a significant factor in the regulation of the activities of heme-containing enzymes. In the context of aging, a long-term decrease in ALA-S activity would have special significance. A persistent decrease in liver heme levels might impair the ability of aging animals to detoxify environmental poisons (cytochrome P-450), synthesize important unsaturated lipids (cytochrome b_5), or dissipate peroxides (catalase). Alternatively, a decrease in liver ALA-S activity might cause a redistribution of whole body heme to satisfy crucial liver requirements. In order to clarify these aspects of heme metabolism, we began to define the changes that occur in the regulation of heme production, utilization, and degradation in the aging rat.

δ -Aminolevulinic Acid Synthetase Activity During Aging

In these studies, the activity of the enzymes was determined either in tissue homogenates, or isolated mitochondria from rats of increasing age. As seen in Table I, both the basal and ethanol-induced activities of hepatic ALA-S showed a highly significant decrease with increasing age.

By comparison with the values for hepatic ALA-S, the activity of this enzyme in other tissues exhibited different patterns of regulation (Table II). It is clear that the ability of the activity to respond to induction by alcohol was tissue-specific. In addition, tissue specificity was also seen in response to nutritional status (30). Moreover, although a significant age-related decrease in enzyme activity was seen in the liver, heart and brain, it was not apparent in the kidney. These observations raise important questions regarding the nature of the control of ALA-S activity in mammalian tissues (30). However, the critical observations are that a significant age-related decrease did occur in the liver, the major site of heme synthesis, and that this pattern was not seen in the kidney. It should, therefore, be possible to design a study in liver and kidney, where the activities of heme-containing proteins are compared as a function of age. Furthermore, heme catabolism by the major pathway could also be examined. In this manner, activities such as catalase, cytochrome P₄₅₀ and mitochondrial cytochrome levels, and heme oxygenase activity would be compared in tissues in the presence or absence of age-related alterations in ALA-S activity. Our laboratory has recently completed the first of such

TABLE I

δ -Aminolevulinic Acid Synthetase Activity in Liver Mitochondria (nmol ALA formed/h/g protein)^a

Age (months)	Saline	Ethanol	Ethanol/saline
1	322 $\pm$ 40	961 $\pm$ 225	3.0
12	151 $\pm$ 29 (-53%) P < 0.005	271 $\pm$ 46 (-72%) P < 0.02	1.8
24	69 $\pm$ 16 (-54%) P < 0.05	188 $\pm$ 49 (-31%) NS	2.7

^a Food was withheld for 24 hours prior to sacrifice. Ethanol was administered by intraperitoneal injection at a dose of 300 mg/100 g body weight 4 hours prior to sacrifice. Saline-treated animals received an equivalent volume of 0.9% NaCl. ALA synthetase activity was determined as described in reference 30. Values are the mean $\pm$ SEM of triplicate determinations on at least 6 animals. P values are obtained using Student's t-distribution. All values are compared to those directly above them. NS = not significant at P > 0.05.

TABLE II

δ-Aminolevulinic Acid Synthetase Activity in Kidney Homogenates (nmol ALA formed/h/g protein)^a

Age (months)	Saline	Ethanol	Ethanol/saline
1	98 ± 18	119 ± 29	1.2
12	77 ± 41 (-21%)	143 ± 30 (+20%)	1.9
24	NS 105 ± 38 (+36%) NS	NS 173 ± 33 (+21%) NS	1.6

δ-Aminolevulinic Acid Synthetase Activity in Heart Homogenates (nmol ALA formed/h/g protein)^a

Age (months)	Saline	Ethanol	Ethanol/saline
1	55 ± 13	61 ± 15	1.1
12	31 ± 9 (-44%)	31 ± 9 (-49%)	1.0
24	NS 7 ± 6 (-74%) P < 0.05	NS 9 ± 7 (-71%) NS	1.1

δ-Aminolevulinic Acid Synthetase Activity in Brain Homogenates (nmol ALA formed/h/g protein)^a

Age (months)	Saline	Ethanol	Ethanol/saline
1	92 ± 18	59 ± 11	0.6
12	39 ± 8 (-58%) P < 0.05	37 ± 8 (-37%) P < 0.2	0.9
24	16 ± 6 (-59%) P < 0.05	14.5 (-54%) P < 0.05	0.9

^a Animals were treated as described in Table I. ALA synthetase activity was determined as described in reference 30. Values are the mean ± SEM of triplicate determinations on six animals. All values are compared to those directly above them. P values from Student's t-distribution given in all cases even if >0.05.

studies (31), the data from which are summarized below.

Mitochondrial Cytochromes and Succinic Dehydrogenase Activities

In Figure 3, the liver and kidney levels of a heme-containing (cytochrome oxidase) and a nonheme containing (succinic dehydrogenase) respiratory chain activity are compared. It is clear that no significant age-related changes occurred in either the amount or the activity of liver cytochrome oxidase, while the kidney activity decreased significantly during the 12-24 month period. By contrast, succinic dehydrogenase activity in both tissues was altered, and displayed a 70% decrease in the aged animal. No significant changes were observed in the levels of other mitochondrial cytochromes (31).

Microsomal Heme Metabolism

Age-activity relationships for cytochrome P₄₅₀ and heme oxygenase are shown in Figure 4. Hepatic cytochrome P₄₅₀ levels increased during growth, and exhibited no decline in aging. It should be noted that kidney P₄₅₀ levels could not be adequately quantitated in this study and have been omitted. The activity of microsomal heme oxygenase actually increased during development and aging in the liver.

Catalase Activity During Aging

When examined in liver and heart, catalase activity increased progressively with age so that the levels in the 24-month old were at least twice those seen in the 2-month old (Fig. 4). Kidney catalase activity followed a different pattern that peaked in the adult and declined in the aged animal.

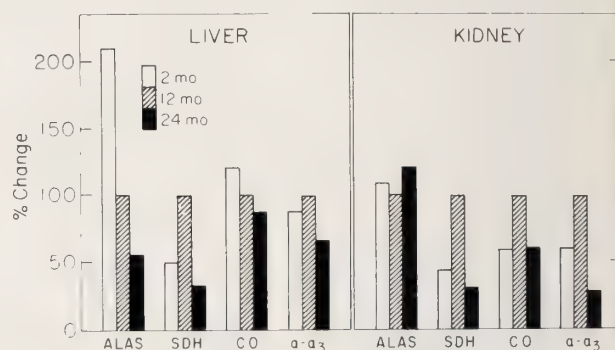


FIG. 3. Cytochrome oxidase activity, cytochrome a-a₃ content, and succinic dehydrogenase activity in liver and kidney in mitochondria isolated from young, adult, and aged rats. Enzyme activities and cytochrome levels were determined as described in reference 31. ALA-S data is that for uninduced animals as described in Tables I and II.

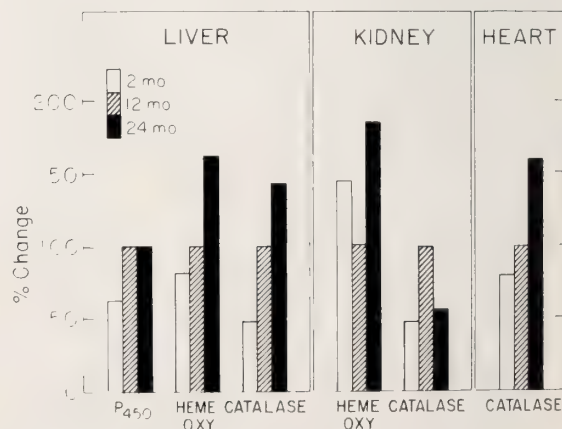


FIG. 4. Cytochrome P₄₅₀ levels, heme oxygenase activity, and catalase activity in young, adult, and aged rats. Parameters in liver, kidney and heart tissues were determined as described in reference 31.

Evaluation: The results of this study and the previous data (Tables I and II) form the basis for the following general scheme of heme metabolism during aging. In the liver, there occurs a progressive decrease in both basal and ethanol-induced activities of ALA-S. This age-related phenomenon is seen in heart and brain tissue but is absent in the kidney. In addition, each organ displays an activity pattern which indicates that tissue-specific regulation is occurring. These results indicate that the flux through the heme biosynthetic pathway may decrease in a time-dependent manner. By contrast, an examination of several pathways for heme utilization, which exist in various cellular compartments, indicate no coordinated decrease in heme flux. Furthermore, the principal pathway for heme catabolism remains unimpaired during aging.

These observations illustrate that the relationship of ALA-S activity in liver mitochondria to overall hepatic heme-protein metabolism is complex. In addition, they indicate that further detailed studies are necessary to develop the major aspects of heme metabolism during aging. Some areas for further inquiry could include: 1) quantitation of actual heme production rates in aged animals, 2) measurement of levels of other heme-proteins (for example, cytochrome b_5), 3) Establishment of values for total heme degradation, 4) Estimation of heme flux to the liver from other tissues. In this last area, it has recently been suggested (32) that exogenously-supplied heme can equilibrate readily with the pool that furnishes the heme for cytochrome P_{450} . Hence, the net amount of heme available to the liver may be larger than the hepatic production. Such a pathway may assume special importance if the rate of liver heme production decreases dramatically with aging.

Mechanisms of Regulation of ALA-S

While the studies already presented have described important phenomena that relate to the metabolism of heme in the aging organism, the questions of mechanism at the molecular level are yet a far frontier. In order to establish the molecular basis for the regulation of ALA-S, and to understand its role in aging, we have purified the mitochondrial enzyme from the livers of young uninduced rats (13). This purified preparation consisted of an apparently homogeneous protein of molecular weight 58,000, that exists as a dimer in the native state. Antibodies prepared against the purified enzyme were monospecific, as judged by diffusion analysis on an Ouchterlony plate (33). Using immunoelectrophoresis, the amount of ALA-S protein

has been measured in detergent-solubilized mitochondria isolated from rats treated with porphyrinogenic agents. The results indicate that in addition to changes in the amount of ALA-S protein, regulation of pre-formed enzyme, over a wide range of activities, also occurred (33). The control of pre-formed enzyme activity is consistent with preliminary studies in aging animals (Table III). It is clear that extraction of mitoplast preparations derived from aged animals with the nonionic detergent Lubrol WX, unmasks ALA-S activity. The activity of the solubilized enzyme is equivalent in preparations derived from rats of widely differing ages despite the marked age-related decrease in activity observed in intact mitochondria. Parallel observations were first made when activity in Lubrol WX extracts of untreated and induced rats were compared and found to be identical (11). Antibody titration studies currently in progress should further define the role of activity regulation in this phenomenon.

Perhaps, one mechanism by which ALA-S activity is regulated is through its association with an activator protein present in close association with the catalytic subunit of the enzyme. Such a protein has recently been isolated in this laboratory (33, 34). Partially purified preparations of the activator protein were able to stimulate the activity of soluble ALA-S *in vitro* over a 3-4 fold range. Indeed, we have postulated that the mechanism of action of the activator may be to cause the formation of high molecular weight aggregates of ALA-S with greater catalytic activity. An alternative explanation might be that the activator protein affects the binding of the catalytic ALA-S dimer to the mitochondrial inner membrane in a manner that results in a greater rate of activity. During aging, the association of ALA-S with the membrane may be altered such that a lowered catalytic activity results. Further studies on the nature and interactions of the activator with the catalytic protein may provide important insights into the regulation of heme production during both aging and porphyria.

TABLE III

δ -Aminolevulinic Acid Synthetase Activity in Isolated Mitochondria and Lubrol-WX Extracts of Mitoplasts (nmol ALA formed/h/mg protein)^a

Age months	n	Mitochondria	Lubrol-WX Extract
1	(8)	0.32	2.5
12	(6)	0.15	1.8
24	(7)	0.069	2.6

^a Mitochondria and lubrol-wx extracts of mitoplast preparations were obtained from 24-hour-fasted rats as described in detail in reference 13. ALA-S activity was determined as described in reference 13.

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Age-related Defects in Mammalian Myocardium: Effects of Dexamethasone

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Abstract

In a comparative study of the mechanical and biochemical responses of the myocardium of young, and aged cats, it was observed that contractile tension decreased and the velocities of contraction and of relaxation declined with age. Concomitantly, the activity of the adenylate cyclase in the tissue decreased. It was also found that while twice the normal amount of calcium concentrations were required to elicit a positive force-frequency in the aged myocardium, a positive force-frequency response was observed in myocardia from young animals at all concentrations of calcium studied. Pretreatment of the aged cats for three days with 1 mg/kg dexamethasone reversed all the above age-related alterations.

A number of changes in the mechanical responses of the mammalian myocardium have been associated with aging. It has been shown that the resting tension increases (1) and there is evidence that the aged myocardium may not generate as much tension as the young and adult muscle. The time course of contraction, and especially of relaxation, is prolonged in the aged tissue (2). The resting and active stiffness of the heart also are shown to increase with aging, while the velocity of shortening decreases (3). The mechanism of these changes in the aged myocardium is not well understood. We have been interested in the mechanism of the inotropic action of dexamethasone in cardiac muscle, which we have previously reported (4,5). We observed a very significant effect of dexamethasone treatment on the peak isometric tension, as well as in the velocities of contraction and of relaxation, in papillary muscle preparations from the dog, the cat, and the rabbit. We also observed that the inotropic effect of dexamethasone treatment was not via the stimulation of the catecholamines; since in the reserpinized animals, dexamethasone treatment over-

came the negative inotropic effects of catecholamine depletion (5), which were observed in the control reserpinized animals. The response to dexamethasone treatment suggests that this compound may have multiple-site effects.

In the present paper, we are reporting on the mechanism of inotropic effects of dexamethasone on the age-related defects in the myocardium of cats. This study covers some of our findings on young, adult and aged cats. As it will be shown below, dexamethasone significantly reverses the age-related changes in the myocardial sarcolemma and contractile proteins. A possible mechanism of action of the steroid hormones in the maintenance of the integrity of the myocardial functions is suggested.

Materials and Methods

Kittens, young adult cats, and aged cats (eight-years old) were anesthetized with sodium pentobarbital (30 mg/kg). The heart was removed and the right ventricle was incised along the septal margin. The thinnest papillary muscle or a trabeculae was resected after the two ends of the tissue were tied up with a silk surgical thread. The tissue was attached to a transducer immersed in a tissue bath that was continuously perfused with oxygenated Tyrodes solution. The tissue bath was jacketed at 35.0 ± 0.02 °C as was a second bath reservoir. The second reservoir was used to modify the composition of the solution bathing the tissue. The tissues

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Some aspects of this investigation were presented at the 9th annual meeting of the American Aging Association, Washington, D.C.

Some of the components of the computer system used in this work were purchased by a generous gift from Merck, Sharpe and Dohme to Dr. Penefsky.

were stimulated at a rate of 6/min for at least 60 minutes before the recordings began. Control recordings were made at various intervals during the equilibration period. Normally between 30–45 minutes of the incubation, the effects of anesthesia and resection appeared to be over, and a steady recovery of the response reached a steady-state.

Experimental Procedures

The isometric tension developed by the muscle as well as the differential of this signal were simultaneously displayed, in parallel, on the screen of a model 564 Tektronix oscilloscope. The differentiated signal was also digitized and the data was stored in the memory of a Wang programmable calculator. The digitized data were analyzed for the points of interest on the curve of the contractile tension and, thus, beat by beat analysis of the curve of the isometric tension could be achieved, when the experimental conditions were altered. In this manner, transient changes as well as steady-state data were obtained for further kinetic studies.

1. Length-tension studies: after the isometric tension at the initial length (L_i) was recorded, the preparation was stretched by an increment of 0.2 mm, and beat by beat changes in the consecutive contractions were recorded until a new steady-state was reached. The procedure was repeated until a maximal, or near-maximal, tension was developed by the preparation. All further studies were done at the length at which maximal tension was developed, defined as the optimal length (L_o).

2. Force-frequency studies: A relationship was obtained in the preparations between the frequency of stimulation and the mechanical response of the muscle preparation. The initial rate of stimulation (6/min.) was stepwise increased to 60/min., and beat by beat recordings were made during the transient state at each frequency, until a new steady-state was reached.

3. Acceleration: In addition to the stepwise increase in the frequency of stimulation as mentioned above, the rate of stimulation was increased tenfold in one step (from 6/min. to 60/min.), and beat by beat recordings were made until a new steady-state was reached.

4. Deceleration: after the preparation had reached a steady-state at high frequency of stimulation, the rate of stimulation was suddenly reduced to a minimum (6/min.), and beat by beat recordings were made until a new steady-state was reached.

5. Electrophysiologic Studies: In a group of cats of various ages, transmembrane action potentials

were simultaneously recorded with the mechanical response. Standard glass microelectrodes were filled with 3M KCl. Action potentials were recorded in control preparations, and in preparations pretreated with various compounds (see below), for observations of possible changes in the contour of the action potential.

Treatment of the Animals

Cats were treated with reserpine (5 mg/kg IM) to deplete the heart of catecholamines. Twenty-four or forty-eight hours later, the animals were sacrificed, a papillary muscle was removed for the mechanical and electrophysiologic studies, and the rest of the ventricle was quickly frozen for fluorometric measurement of tissue catecholamines.

Cats of various ages were pretreated with dexamethasone (1mg/kg, I.M.), for three consecutive days and, on the fourth day, they were treated with 5 mg/kg of reserpine. The ventricular muscle from these cats was also immediately frozen for catecholamine studies.

Cats were pretreated with three injections of dexamethasone as mentioned above, and were studied within a day or two after the last injection.

In a group of control and dexamethasone-treated cats of various ages, following removal of the papillary muscle, the rest of the ventricle was homogenized, and the adenylate cyclase of the homogenate was assayed (6).

The preparations were subjected to Tyrodes solutions containing calcium concentrations ranging from 5.4 mM (twice normal concentration), to 0.675 mM (1/4 normal concentration), by exchanging one solution with the other. The effects of calcium concentration on the mechanical and electrophysiologic responses of the myocardial preparations were recorded.

Analysis of the Data

The data generated by the transducer and digitized were programmed for on-line analysis of the isometric tension developed at various experimental procedures. Contractile tension during the early and latter phases of contraction, peak contractile tension, tension in the muscle during early and late stage of relaxation, as well as the time course of various phases of contraction and relaxation were tabulated for each consecutive contraction. Besides the above data, the average velocity during the early and late phases of contraction and relaxation were calculated and printed out.

Results

Length-tension responses: When comparable preparations from kittens, adult and aged cat myocardium were stretched between Li and Lo (methods), contractile tension developed for relative lengths of the preparations appeared to be slightly higher in the kittens than in the young adult animals, and significantly higher than in the old preparations. Resting tension, at the Lo observed in the young preparations was about 50% of the peak contractile tension; while in the old myocardial preparations, resting tension at Lo was equal, or even higher than the active tension. In the adult preparations, the value was intermediate between the young and the old. There was a large variation of resting tension in preparations of the same group.

Figure 1 illustrates peak contractile tension as recorded at steady-state from a group of young, adult, and old cats, at various frequencies of stimulation. Whereas in the young animals the contractile tension was more than doubled when the frequency of stimulation was increased tenfold, in the old group, there was a slight, but significant decline in the peak contractile tension at high frequency of stimulation. By contrast, the old dexamethasone-treated myocardium did demonstrate a significantly positive force-frequency response (about 40%).

In tissues from young cats the responses to stepwise increases in the frequency of stimulation also included an increase in the velocity of contraction and of relaxation, and an abbreviation of the contraction-relaxation cycle. In the preparation from aged cats, there were small reductions in the contractile tension and the duration of the contraction-relaxation cycle, while the velocities of contraction and of relaxation were not significantly changed. When tissue from a dexamethasone-treated aged animal was studied with the same procedure, there was a slight, but significant increase in the amplitude of the contractile tension as the frequency of stimulation was increased. The velocities of contraction and of relaxation were equally increased when the frequency of stimulation in the old dexamethasone-treated myocardial preparations were stepwise increased.

Acceleration: In a bathing solution containing 2.7 mM calcium (normal concentration), acceleration of the young and adult myocardium consistently increased the isometric contractile tension at consecutive contractions, until a steady-state tension was reached after six to eight contractions. By contrast, in the aged preparations, the contractile tensions elicited were alternating at consecutive

AGING EFFECT ON F-f RESPONSE: EFFECTS OF DEX

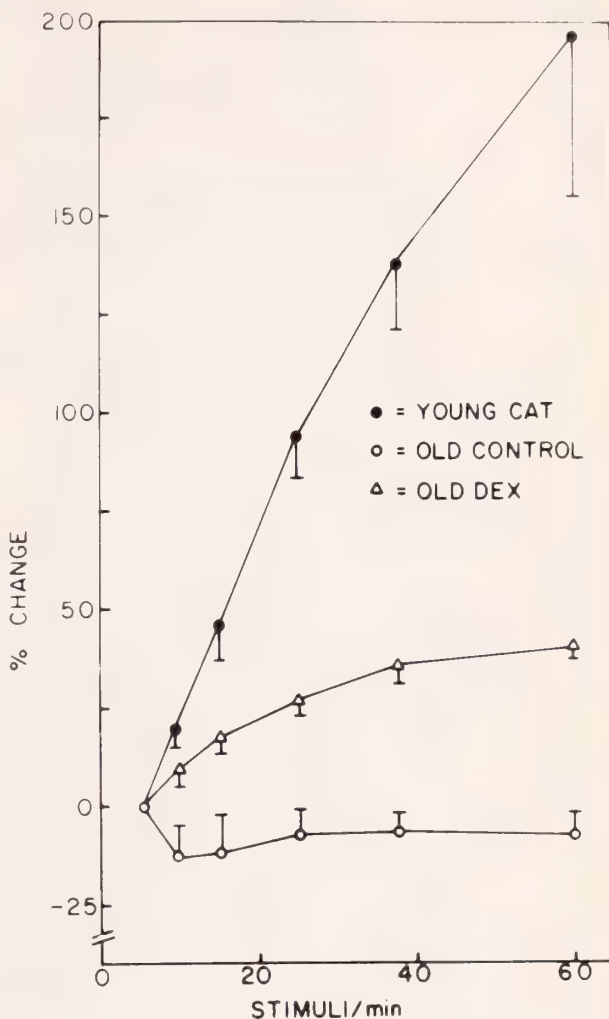


FIG. 1. Percent change in the isometric contractile tension, as the frequency of stimulations was stepwise increased from 6/min to 60/min. Vertical bars: S.E. of Means.

beats, with an overall negative trend. Steady-state tension was not reached before 15–18 contractions. When the old animal was pretreated with dexamethasone, the steady-state response to acceleration was an increase in the peak contractile tension (Table I).

Deceleration: In the young and adult myocardium, reducing the frequency of stimulation induced an initial post-extra systolic potentiation, with consecutively diminishing contractile tensions, until a steady-state was reached, within six to ten contractions. In the old untreated myocardium, post-extrasystolic potentiation was absent, and the following contractions demonstrated increasingly

higher peaks, until the steady-state was reached within 15–18 beats. When old dexamethasone-treated myocardial preparations were decelerated, a post-extra systolic potentiation and a negative going staircase was obtained. This response was qualitatively similar to that observed in the young preparations.

Response to a Gradual Increase in Calcium Concentration: When the bathing solution containing 2.7 mM calcium was switched to one with 5.4 mM calcium, and minute by minute response of the

TABLE I

Steady-state isometric tension before and after acceleration of the heart in bathing solutions containing various concentrations of calcium. Tension: g/mg tissue.

EFFECTS OF RATE AND CALCIUM CONCENTRATION ON PEAK CONTRACTILE TENSION

CALCIUM CONCENTRATION	5.4 mM		2.7 mM		1.35 mM	
	6 min	60 min	6/min	60/min	6/min	60/min
YOUNG HEART	0.5	1.30	0.26	0.77	0.24	0.90
OLD UNTREATED	0.63	0.87	0.30	0.30	0.13	0.14
OLD DEX TREATED	0.83	33	0.55	0.89	0.26	0.77

myocardial preparations was recorded, the following differences were observed (Fig. 2). The young myocardium appeared to respond after a lag time following the switch (first arrow), and the contractile tension developed had a fast rising slope. So much so, that within a minute before the equilibrium was assumed to have been reached in the tissue bath (second arrow), the young myocardium had reached a steady-state. By contrast, the old untreated myocardium demonstrated a slower rate of rise in the peak contractile tension, and the steady-state in the peak contractile tension, was only achieved about 15 minutes after the presumed equilibration in the tissue bath. When an old dexamethasone-treated heart preparation was subjected to the above procedure the response was virtually indistinguishable from the young myocardium. Figure 3 illustrates the steady-state response elicited from a group of untreated and dexamethasone-treated old cats in bathing solutions ranging between 5.4 mM (two times normal concentration), and 0.675 mM (1/4 normal concentration). The average tension developed by the dexamethasone-treated preparations at each calcium concentration

RESPONSE OF CONTROL AND DEX-TREATED 8 YRS CATS AS $[Ca^{++}]$ WAS GRADUALLY INCREASED TWO FOLD

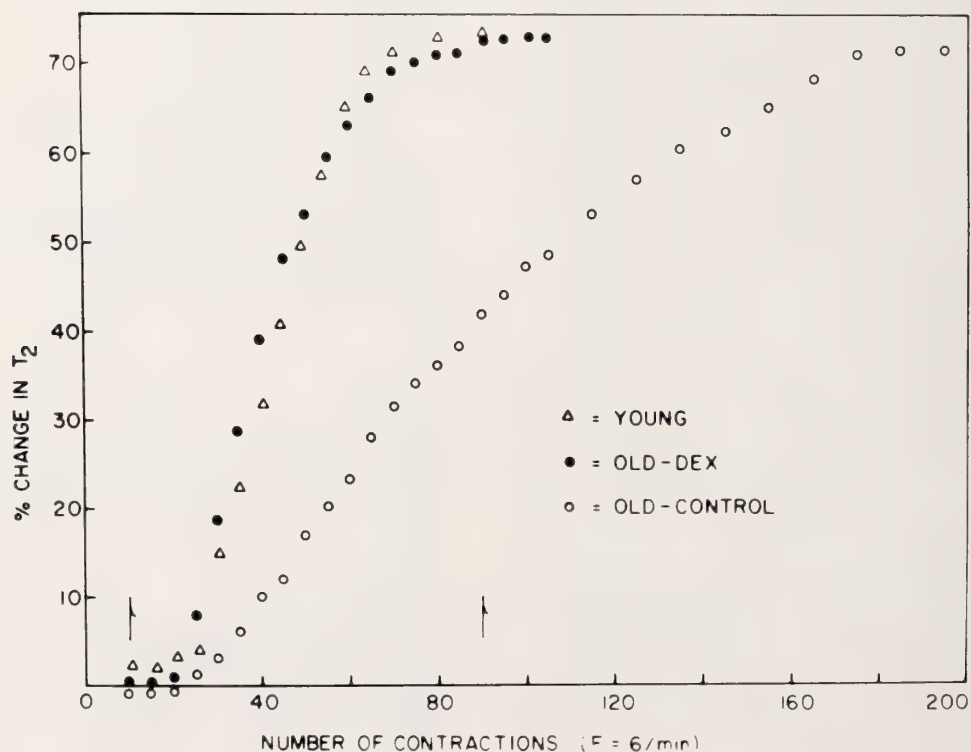


FIG. 2. Percent increase in the isometric contractile tension recorded from a young (Δ), an old untreated ($\circ$), and an old dexamethasone-treated ($\bullet$) cat papillary muscle, as a function of gradually increasing calcium concentration in the tissue bath (see text). First arrow: start of the change in the calcium concentration; second arrow: calcium equilibration in the tissue bath.

PEAK TENSION DEVELOPED AT VARIOUS CALCIUM CONCENTRATIONS

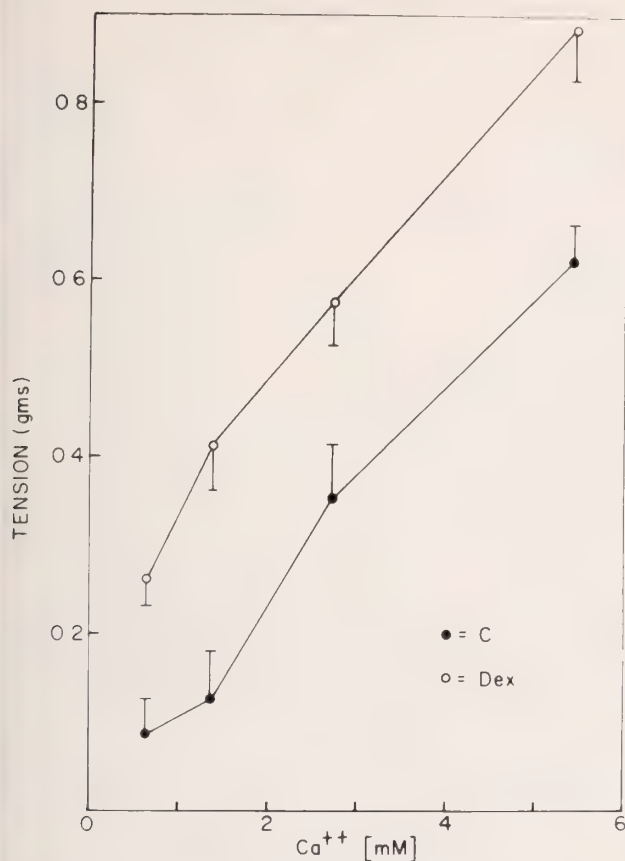


FIG. 3. Steady-state contractile tension recorded from untreated (C), and dexamethasone-treated (Dex) old cat papillary muscles as the calcium concentration in the tissue bath was varied between 2 times normal concentration and 1/4 normal concentration. Vertical bars: S.E. of the Means.

was significantly higher when compared to the untreated preparations, however, percent increase in tension appeared to be significantly higher in the untreated myocardial preparations between normal concentration of calcium, and two times normal concentration. While the dexamethasone-treated preparations increased contractile tension 50% when the calcium concentration in the bathing solution was doubled, the untreated preparations increased peak tension almost 80%. By contrast, when the calcium concentration in the bathing solution was reduced below normal concentration, contractile tension fell to a larger extent in the untreated preparations. In a bathing solution with calcium 1/4 normal, peak contractile tension developed by the untreated preparation was only 23% of that in normal concentration; while in the dexamethasone-treated preparations it was 45% of peak tension in normal Tyrodes solution.

Table I is an illustration of the response of young adult and old myocardial preparation to acceleration in the presence of various calcium concentrations in the bathing solution. In the young cat myocardium, a positive response to acceleration was present at all concentrations of calcium in the bathing solution. Only at high concentrations of calcium (two times normal) did the old untreated preparation demonstrate a positive acceleration; while at normal and half normal calcium concentrations, there was no positive response to acceleration. By contrast, in the dexamethasone-treated old preparation there was a positive acceleration response at all calcium concentrations in the tissue bath.

Electrophysiologic Studies: Figure 4 illustrates a comparison made between the mechanical response and the contour of the transmembrane action potential recorded from a control adult cat papillary muscle (Fig 4A) and preparations pretreated, as indicated in Methods. Panel B in Figure 4 represents the recordings from a reserpinized cat myocardium. It can be seen that the amplitude of the contractile tension curve is highly depressed in this preparation, the velocity of contraction and of relaxation, as indicated by the dT/dt curve, is also flattened, indicating a decline in the rate of contraction and relaxation. However, the contour of the action potential in this preparation is minimally different from the control preparation (compare panels A and B). Panel C demonstrates the electrical and mechanical response of the preparation shown in panel B, following an increase in the calcium concentration in the bathing solution to 5.4

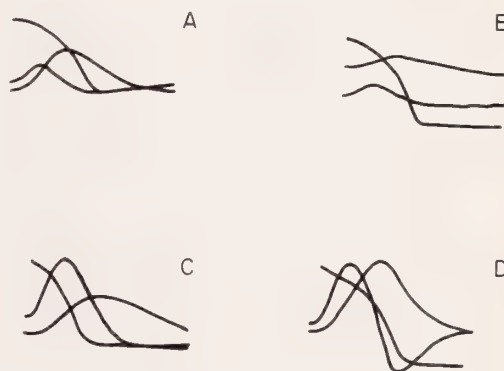


FIG. 4. Transmembrane action potential, isometric tension (monophasic curve), and dT/dt (biphasic curve) recorded from cat papillary muscles under various experimental conditions. A: Control muscle in normal Tyrodes solution; B: reserpinized muscle in normal Tyrodes solution; C: reserpinized muscle in a Tyrodes solution containing two times normal calcium concentration; D: Papillary muscle from a cat pre-treated with dexamethasone, and subsequently reserpinized (see methods) in normal Tyrodes solution.

mM. Whereas the amplitude of the contractile tension and the rate of tension development were increased, and the duration of the action potential was typically shortened in the presence of high calcium, there was no change in the rate and duration of relaxation. In panel D is shown a recording from a cat myocardium pretreated with dexamethasone, and then reserpinized (see Methods). Not only are the amplitude of tension and the velocities of contraction and of relaxation significantly higher than the control (Panel A), but the contour of the action potential is also different from that of a dexamethasone-treated myocardium. When the homogenate from the ventricles of preparations illustrated in panels B and D were analyzed for catecholamine content, neither one had measurable levels.

Biochemical Studies: Because the experiments with reserpinized and dexamethasone-treated reserpinized preparations did not indicate that dexamethasone may have stimulated the catecholamine effect, in a preliminary study, we compared the activity of the adenylate cyclase in the ventricular homogenate, with the mechanical response elicited by the respective papillary muscles. Cats of various ages were used in this study, and the results are illustrated in Figure 5. We observed a direct relationship between the amplitude of isometric tension, the velocity of contraction and of relaxation, with the level of cyclic AMP assayed in the homogenate of the respective hearts. Furthermore, it became evident that the younger hearts developed higher contractile tension, and were higher in adenylate cyclase activity as compared to the older cats.

Because the data mentioned above suggested an age-related decline in the mechanical response of the myocardium and the activity of the enzyme adenylate cyclase in the ventricular homogenate, we selected three groups of cats of various ages (three months, one to two years, and eight years) for a controlled study. Cats in each group were closely matched in age; and in each group, some were used as control and others were treated with dexamethasone. Data from such experiments are presented in Table II. It can be seen that in the kittens, the basal level of the cyclic AMP assayed was highest, as compared to the older groups. Dexamethasone treatment did not appear to have an effect in the activity of the adenylate cyclase. Furthermore, none of the homogenates in this group was stimulated by the addition of epinephrine. Peak contractile tension developed by all the kittens was high, and so were the velocities of contraction and of relaxation, not shown in the Table II.

RELATIONSHIP BETWEEN MECHANICAL PERFORMANCE AND ADENYLATE CYCLASE ACTIVITY OF HEART MUSCLE

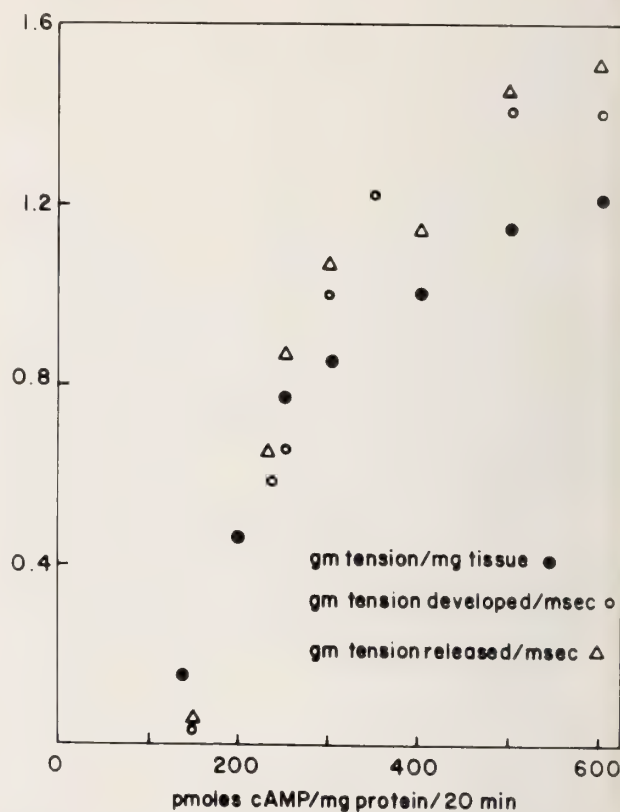


FIG. 5. Isometric contractile tension (●), the velocity of contraction (○), and of relaxation (△) recorded from cat papillary muscles, and their respective activity of adenylate cyclase, as measured from the cyclic AMP generated in the ventricular homogenate.

In the adult group, although the basal level of the cyclic AMP was not significantly greater in the dexamethasone-treated preparation, it was highly stimulated by the addition of epinephrine (Table II). As a result, the total level of cyclic AMP in the dexamethasone-treated preparation was significantly higher than in the untreated homogenate. In effect, the total cyclic AMP content of the adult dexamethasone-treated homogenate was comparable to those of the kittens. In the adult dexamethasone-treated papillary muscle, contractile tension also was comparable to those of the kittens, while in the untreated myocardium contractile tension was significantly lower.

In the aged animals, the effects of dexamethasone treatment appeared to be most significant. The basal level of the cyclic AMP in the dexamethasone-treated aged myocardium was comparable to those in the kittens (see Table II). In spite of an epineph-

TABLE II

A comparison of the basal and epinephrine-stimulated activity of the adenylate cyclase from the ventricular homogenate and the isometric contractile tension from the respective papillary muscle. In each group effects of dexamethasone treatment on the above parameters is compared to the controls.

Condition	Age	cAMP: pmoles/mg tissue/min.			γ /mg tissue Tension (max)
		Basal	Epi-stimulated	Total	
Untreated	3 mos	34.3	0	34.3	1.380
"	"	40.7	0	40.7	1.120
"	"	33.4	0	33.4	0.910
dexa-treated	"	25.8	0	25.8	1.56
untreated	adult	13.1	0	13.1	0.530
dexa-treated	"	17.3	18.5	35.8	1.01
untreated	aged	4.4	9.0	13.4	0.11
dexa-treated	"	31.1	0	31.1	0.58

rine stimulation in the untreated homogenate, the total level of the cyclic AMP assayed in the dexamethasone-treated homogenate was more than two times greater than in the control. Although the relative contractile tension in the dexamethasone-treated papillary muscle was significantly higher than that in the control, when compared to the adult heart, it was only comparable to the untreated adult myocardium.

Discussion

The experiments shown above were focused on the possibility of multiple site defects in the aging heart, as well as on the possible mechanisms of action of dexamethasone in reversing the age-related alterations. The alterations illustrated in the above experiments clearly demonstrate that the cardiac sarcolemmal properties change, as the animal ages. Electrophysiologic changes of the sarcolemma, in the course of development have been demonstrated in many laboratories. Although many of these changes take place in the embryonic stages,

as reported by other investigators (7-9), these changes do not stop as the animals become mature and age. On the contrary, these alterations appear to be an on-going process throughout the life span of the animal, as shown by Rosen et al (10).

The contractile elements equally undergo age-related alterations. This is readily indicated by the decline in the rate (velocity) of the tension developed, as well as the peak tension generated by the myocardium. Another striking change in the aging myocardium is the rate at which it relaxes. The time course of relaxation is greatly prolonged, and the velocity of relaxation is decreased in the aged myocardium. Thus, not only is the actomyosin interaction altered in the course of aging, but the sarcoplasmic reticulum also appears to be affected in the old myocardium.

Whether the multiple site defects in the course of aging stem from the sarcolemmal alterations, or if the alterations described in the contractile elements are from unrelated origins cannot be speculated from our data, although all the phenomena studied in this work appear to be reversed by dexamethasone treatment. Although the biochemical data are

highly suggestive of an age-induced defect in the myocardium, the environmental effects should also be highly important factors in the degeneration of the myocardial functions. For instance, the disease-related alterations, metabolic defects, nutritional deficiencies, anoxia, fatty infiltration, to name a few causes, may play significant roles in the function of the myocardium. However, in species that have been raised under highly controlled environments such as rats, still the age-related defects in the myocardium and the reversal of these defects following treatment with dexamethasone can be observed (unpublished data).

The results of the experiments with dexamethasone appear to imply that perhaps the age-induced alterations observed above, have a common denominator; since a brief treatment of the aged animals with the hormone appears to significantly reverse the mechanical and the biochemical alterations in the myocardium. In this respect, although the data shown for the old cats (Table II) indicated a high level of cyclic AMP and a low contractile tension in the dexamethasone-treated myocardium, it should be realized that the myocardial tissue obtained from old animals is usually infiltrated with fat tissue, if not partially infarcted. Whereas the myofibers are not expected to regenerate following dexamethasone treatment, the level of the cyclic AMP in the viable tissue may increase. Therefore, it is not surprising that in such animals, the papillary muscle preparations are relatively hypotonic despite a high cAMP content of the homogenate. Because, in effect, the homogenate is presumably provided from viable tissue.

Besides the stimulation of the adenylate cyclase, dexamethasone also appears to replenish the calcium stores of the myocardium. This effect becomes evident from the calcium titration experiments (Fig. 3), when it was shown that the contractile tension in the untreated old myocardium became extremely sensitive to the concentration of calcium in the bathing solution, whereas in the dexamethasone-treated old hearts, the effects of concentration of calcium in the bathing solution on the contractile tension developed by muscle were not as highly dose dependent.

The difference between the young and old myocardium in the response to a gradual increase in the ambient calcium concentration (Fig. 2), suggests that perhaps, in the young animal either there were more calcium binding sites available on the sarcolemma or, alternatively, the same number of sites had higher binding capacity. Whatever the reason, dexamethasone treatment of the old animal restored the mechanical responses of the myocardium

(Table I). On the sarcolemmal basement membrane, a glycoprotein structure: glycocalyx, with high sialic acid content has been identified (11,12). The polyanionic sites on the glycocalyx have been suggested as the calcium binding sites (13,14). Whether dexamethasone stimulates the glycocalyx remains to be seen.

Another observation supporting the suggestion that the dexamethasone treatment enhances the replenishment of the myocardial calcium stores is illustrated in Figure 3. The old dexamethasone-treated myocardium appeared to have access to larger stores of calcium, when compared to the untreated myocardium.

The force-frequency response of the old untreated and dexamethasone-treated myocardial preparations also indicated that in the untreated myocardium, perhaps the calcium stores could not be replenished as readily as in the dexamethasone-treated preparations. Therefore, at calcium concentrations of 2.7 mM, and below, as the interval between the consecutive stimuli shortened (frequency of stimulation increased) a deficit in the supply of calcium appeared, which resulted in negative inotropism (Table I, middle row), while the young and the dexamethasone-treated old myocardia could generate higher tensions as the frequency of stimulation increased, regardless of the ambient calcium concentration. A supportive finding to this suggestion was that at high ambient calcium concentration: 5.4 mM, even the old untreated myocardium could generate a positive response at higher frequency of stimulation (Table I).

Although the sarcolemmal properties of the myocardium may be an important feature of the aging heart, other changes in the contractile properties of the old myocardium have also been reported (15, 21).

Our studies strongly suggest that dexamethasone may mimic a natural control of the adrenal steroid hormones on the integrity of the myocardium. The role of steroid hormones on the induction of proteins and enzymes has been demonstrated in several laboratories (16-23). It has been reported that 3H-dexamethasone injected in rats is bound to a specific cytosolic protein in the myocardial fibers (20). It has also been claimed that dexamethasone, bound to the receptor protein is transferred to the nuclei of the myocardial fibers (21). It is thus conceivable that dexamethasone shares the properties of the natural steroid hormones and does stimulate the synthesis of various key proteins and enzymes responsible for the maintenance of a normal function in the myocardium. It can be speculated that in the process of aging, either the normal level of

circulating adrenal steroid hormones falls, an effect claimed by some investigators (24), or, alternatively, the sensitivity of the myocardial fibers to a normal level of circulating hormone is reduced. In either case, the effect may be a reduction in the myocardial response and, consequently, a decline in the production of the enzymes and other proteins that are responsible for the maintenance of structural and functional integrity of the myocardium. This suggestion is supported by a recent report, that in the aged animals the synthesis of myocardial proteins is reduced (17, 19).

One of the striking effects of dexamethasone treatment is the rate of relaxation of the myocardium, which may reflect the rate of calcium sequestration by the sarcoplasmic reticulum. In this respect, dexamethasone mimics the role of epinephrine. It is well documented that epinephrine abbreviates the relaxation time, and increases the rate of relaxation. As it was shown in Figure 4, reserpini-zation significantly reduced the rate of relaxation (panel B) and the addition of calcium (panel C), was not effective in reversing this inhibition. However, in the presence of reserpine, a dexamethasone-treated myocardium maintained a fast rate of relaxation (panel D). From our data, it is not possible to suggest whether the effect of dexamethasone treatment is directly on the sarcoplasmic reticular protein, or the consequence of the increase in the level of cyclic AMP, which may intervene in the phosphorylation of the protein kinase.

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Thyroid Function and Dysfunction in the Elderly: A Brief Review

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Abstract

The anatomy, physiology and function of the aging thyroid gland are presented. Hyperthyroidism, hypothyroidism, thyroid neoplasia, and thyroiditis in the elderly are discussed with specific reference to differences between clinical presentation and therapeutic approaches in older versus younger patients.

Introduction

The past two decades have seen tremendous strides made in the area of endocrinology. With the advent of the technique of radioimmunoassay, it has become possible to directly measure aspects of endocrine function that previously could only be assessed by cumbersome and sometimes hazardous indirect studies. These breakthroughs have led to a more complete and precise understanding of thyroid function and thyroid disorders.

As life expectancy has increased, the medical profession has found itself dealing more and more with specific variations of medical problems that apply primarily to elderly individuals. A large literature concerned with differences in thyroid physiology, function and disease between younger and older individuals has been published. This paper summarizes some of the clinically important and useful information that has evolved. Needless to say, many questions regarding thyroid function and dysfunction in the aged remain unanswered. For example, why do so many differences exist between male and female thyroregulatory mechanisms? As techniques become even more sensitive and larger numbers of elderly patients are investigated, answers to many of these questions should be forthcoming.

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Anatomy

As aging takes place, anatomical changes in the thyroid gland may occur. These include atrophy and fibrosis which may result in a decrease in the weight of the gland. The thyroid may become more nodular. Microscopically, nodularity and cellular infiltrates become more apparent. The size of the follicles decreases as does the amount of colloid within them. Interfollicular connective tissue increases (1, 2).

Some of the changes in the thyroid associated with aging may be autoimmune in nature. Both antithyroglobulin and antimicrosomal antibody titers show an increase with age. By the eighth decade, nearly 20% of these patients have elevated serum titers (3). The increase in frequency with aging for both types of antibodies is more pronounced in women than in men.

Physiology and Function

Thyroid function, physiology and regulatory mechanisms in the elderly are comprehensively reviewed by Ingbar (4, 5) and by Sawin et al (6).

There remains considerable controversy regarding whether or not thyroid function becomes abnormal as aging takes place. There is data to suggest that daily iodide turnover decreases with age. On the other hand, the 24-hour radioactive iodine uptake does not change significantly with aging (7). These facts indicate that in the aging thyroid, there is a diminution in the absolute rate of thyroid iodine accumulation. This would imply that the aging thyroid has a diminished rate of hormone forma-

tion. This, however, may not mean that there is a primary defect in thyroid function. The decrease in plasma total and free Thyroxine (T4) concentrations seen in primary thyroid failure does not occur with aging, suggesting that the apparent decrease in thyroidal iodide accumulation seen in aging is a mechanism to compensate for a decrease in the rate of peripheral thyroid hormone degradation (5).

The functional reserve of the aging thyroid gland has been tested by stimulation with exogenous thyroid stimulating hormone (TSH) followed by measurement of serum T4. Unfortunately, some studies demonstrate that the functional reserve is decreased, while others do not. (5). Looking at the same question of TSH-releasing hormone (TRH) testing, the available data suggests that thyroid responsiveness to TSH is not decreased with aging and that it may be increased in males (8).

Some studies show that serum concentration of T4 in the elderly is not affected by age (9) while other studies show the T4 concentration to decrease by approximately 20% (10-12). There is one study that indicates this decrease occurs only in women (13).

Kinetic studies of peripheral T4 metabolism in men demonstrate that the metabolic clearance rate of T4 decreases with advancing age. By the ninth decade, the metabolic clearance rate may be half that measureable in younger men. Interestingly, no change in metabolic clearance rate of T4 has been demonstrated in women (14-16). Serum concentrations of free T4 (9, 11-13) and thyroxine-binding globulin (TBG) (9, 12) remain essentially normal with aging.

It has become evident that triiodothyronine (T3) is the major active thyroid hormone involved with peripheral metabolism. In general, most of the serum T3 is not secreted directly by the thyroid gland, but is derived from peripheral monodeiodination of T4. Apparently, euthyroid individuals' serum T3 concentrations decrease with increasing age (10-12, 17-19). The magnitude of the decrease varies from study to study and one report indicates the decrease is seen only in aging males and not in females. Another recent report illustrates that in patients with normal serum TSH concentrations, the lower limit of serum T3 concentration decreased with advancing age, but in men, the decrease in the lower limit of total T3 values was not notable until after 70 years of age, and in women not until after 80 years of age. Of greatest interest was the finding that the upper limit of T3 concentration in serum was about the same in older and younger individuals (6).

Since serum T3 concentrations appear to decrease in patients undergoing acute or chronic illnesses (10, 20), caloric deprivation or stress (21, 22), one must wonder whether the lower serum T3 levels reported by some observers in aging reflect merely the changes associated with systemic diseases in the aged or represent a real age related alteration in thyroid hormone metabolism (5). The clinical importance of the changes in serum T3 concentrations will be discussed in the Section on hyperthyroidism.

There is data indicating that T3 production rates are lower in older men than in younger men by approximately 25%. This is essentially the percentage by which the T4 disposal rate is decreased in elderly males, suggesting that, in the elderly, the percentage of T4 undergoing degradation through pathways leading to T3 formation is unchanged (14). The definitive studies to elucidate the precise relationships between thyroid hormone metabolism and aging have yet to be reported. The data regarding pituitary-hypothalamic regulating mechanisms in the aged is unclear and frequently contradictory. Most of the evidence to date implies that basal serum TSH concentrations are in the normal range in elderly individuals. There is some evidence to suggest a decreased pituitary responsiveness to TRH in the aging male (18). Overall, the information regarding thyroid function and thyroid hormone metabolism suggests that normal elderly individuals have normal thyroid function and that the decline in metabolic rate seen with advancing age reflects a decrease in functioning tissue mass rather than cellular hypometabolism (5).

Thyroid Neoplasia

Both nontoxic multinodular goiter and toxic nodular goiter occur more frequently with aging (23). Since many thyroid glands seem to contain a solitary nodule on physical examination but are multinodular on direct inspection, the presence of a solitary thyroid nodule in an elderly individual is less likely to represent malignancy than it would be in a younger individual. Still, the usual criteria for suspecting thyroid malignancy such as lack of function, progressive enlargement, hardness and vocal cord paralysis must be applied to solitary thyroid nodules in the elderly as they would be in younger individuals (5). It is apparent that age has an influence on the behavior of thyroid carcinoma (24). Papillary carcinomas, when detected in the third and fourth decades, have a relatively benign course

and longer life expectancy. As a patient with papillary carcinoma ages or when papillary carcinoma occurs in an older individual, it tends to be more aggressive.

Follicular carcinomas that are usually diagnosed in the fifth or sixth decades tend to be more aggressive and have a higher mortality rate than papillary carcinoma. Undifferentiated carcinoma of the thyroid gland usually occurs in patients over the age of 50 years. These tumors are highly aggressive, rarely respond to any form of treatment, and have survival rates of less than one year from the time of diagnosis.

Primary lymphomas of the thyroid gland occur predominantly past the sixth decade, the average age at the time of diagnosis being approximately 60 years (25). They are characterized by a rapidly enlarging neck mass that may be tender. Fifty percent of the patients have hoarseness with or without vocal cord paralysis. Pre-existing lymphocytic thyroiditis may be present. Five year survival rates for patients with primary lymphomas of the thyroid gland vary from 20% to 60%. The histiocytic lymphomas have a worse prognosis than do the follicular, regardless of the modality of therapy. In a series of 60 patients over the age of 50 years with thyroid carcinoma of various cell types, it was found that the ratio of females to males was 5.7 to 1 as opposed to 3.9:1 for patients of all ages (26). Of 185 patients of all ages with thyroid carcinoma, 32% were over the age of 50. In this series, the aggressive nature of thyroid carcinoma in older individuals was reaffirmed by a 25% mortality rate. More than half of the deaths occurred in patients with either papillary or follicular carcinoma. The mean age of patients dying of thyroid tumors is approximately 68 years for both sexes (27).

There is no difference in the therapeutic approach to thyroid carcinoma in younger or aged individuals. Discussion of specific forms of treatment is beyond the scope of this paper but is well covered elsewhere (28).

Hyperthyroidism

Hyperthyroidism in the elderly has been comprehensively reviewed by several authors (5, 29-34). Between 10 and 17 percent of hyperthyroid patients are 60 years or older.

Graves' disease (diffuse toxic goiter) may occur at any age but usually begins in the second, third and fourth decades. In the elderly, toxic nodular goiter is much more common. In thyrotoxic patients over the age of 60 who have thyroidal enlargement,

more than 50% have nodular goiters. Of equal importance, when considering hyperthyroidism in the elderly, is that 20% of the patients have thyroid glands that are not enlarged or not palpable. The typical ophthalmologic findings seen in younger hyperthyroid patients occur less frequently in elderly thyrotoxic individuals.

Unlike the younger thyrotoxic patient who usually presents with a constellation of typical symptoms, the elderly thyrotoxic may present with one predominant symptom referable to a single organ system. The cardiovascular system is the one most frequently involved. Approximately 80% of elderly thyrotoxic patients have cardiovascular abnormalities. The elderly often have underlying cardiovascular disease and the effects of excessive thyroid hormone on the heart, increasing cardiac work and enhancing metabolism by peripheral tissues, can cause development or enhancement of cardiac symptoms. The most common cardiac problems in elderly thyrotoxic patients are palpitations (60%), congestive heart failure (66%) and angina pectoris (20%). Though the cardiac symptoms are frequently caused by accentuation of underlying heart disease, hyperthyroidism can also create cardiac decompensation in patients who have no underlying heart disease (35). Atrial fibrillation is fairly common in thyrotoxicosis especially in the elderly in whom the incidence is approximately 40%. Among older patients with unexplained atrial fibrillation, 10% prove to have thyrotoxicosis. Elderly thyrotoxic patients with atrial fibrillation tend to have slower apical rates than do younger thyrotoxics. They are also less likely to revert to sinus rhythm after returning to euthyroid status. The diagnosis of hyperthyroidism should be suspected in elderly patients with tachycardias or congestive heart failure who do not respond to appropriate digitalization regimens.

Weight loss is a frequent presenting complaint seen in 54%-91% of older hyperthyroid patients. The weight loss seen in younger thyrotoxics is usually associated with increased appetite. Among the elderly, the weight loss may be associated with anorexia. The diarrhea frequently seen in the younger thyrotoxic patient is less common among elderly thyrotoxics. However, the elderly may experience improvement of preexisting constipation. A complaint of heat intolerance is frequent, occurring in approximately 66% of elderly thyrotoxics. Tremor, though frequently present, may be confused with tremors of other causes in elderly thyrotoxic individuals.

Apathetic hyperthyroidism is a form of hyperthyroidism seen almost exclusively in the elderly. Un-

like the classical hyperthyroid patient, the apathetic hyperthyroid is depressed, unanimated and withdrawn. These patients are frequently suspected of having gastrointestinal malignancies or primarily psychiatric disorders. The cardinal features of apathetic hyperthyroidism are 1) elderly individuals with placid, apathetic facies, 2) small goiter, 3) presence of lethargy, depression or indifference, 4) absence of the usual ocular manifestations of hyperthyroidism, 5) substantial muscle weakness or wasting, 6) weight loss, 7) cardiac dysfunction with or without atrial fibrillation (36). Thyroid function studies are usually abnormal though perhaps not as strikingly so as in younger individuals.

Thyroid function studies in younger and older hyperthyroid patients do not differ significantly. The one exception is the serum Triiodothyronine concentration in which the normal range is probably lower in the elderly (10–12, 17–19). T3 thyrotoxicosis may also be more common among older patients. For the above reasons, measurement and proper interpretation of serum T3 concentrations is especially important when thyrotoxicosis is suspected in elderly individuals.

The treatment of choice for hyperthyroidism in the older patient is radioactive iodine. Because of the high incidence of toxic nodular goiters, it has been suggested that the elderly be treated with larger than usual doses of Iodine 131. Because of the possibility of a large outpouring of thyroid hormone due to radiation thyroiditis in the first week after treatment, some authors advocate pretreatment of elderly hyperthyroid patients with antithyroid drugs until they are euthyroid, before treating with radioactive iodine (5). Alternatively, beta adrenergic blocking agents may be used to block peripheral effects of thyroid hormone outpouring. Surgery is rarely indicated in the treatment of older patients with hyperthyroidism.

Hypothyroidism

The major causes of hypothyroidism in the elderly are autoimmune atrophy, Hashimoto's (lymphocytic) thyroiditis, destruction by therapeutic radioactive iodine or surgery, and inherited enzymatic defects (37).

The manifestations of hypothyroidism do not differ substantially between younger and older individuals. However, many symptoms of hypothyroidism are, on the surface, similar to those associated with the aging process alone (cold intolerance, constipation, lethargy, dry skin, cardiomegaly, etc.). This may result in failure to recognize some of the

less advanced cases. Because of underlying cerebrovascular disease in the elderly, they tend to exhibit the neurologic features of hypothyroidism, including dementia and coma, more frequently than do younger hypothyroid individuals (38, 39).

Sawin et al studied 344 relatively healthy people over 60 years of age with TSH, T4, and free T4 index. Twenty-two or 5.9% had clearly elevated levels of serum TSH (>10 uU/ml). This occurred more commonly in women than in men. Ten of the 22 had low serum concentrations of T4 and free T4 index. Another 14% of the 344 subjects had slightly elevated serum levels of TSH ($>5 \leq 10$ uU/ml) but none of them had low concentrations of T4 or free T4 index. It was concluded that the prevalence of hypothyroidism in the elderly is greater than previously thought (6). Others, using TSH as a screening procedure, have confirmed this finding (40). Sawin et al suggest that it may be useful to screen all individuals over the age of 60 years for high serum TSH concentrations as has been done in neonates.

Once suspected clinically, the diagnosis of hypothyroidism in the aged is usually confirmable by laboratory data. Low T4, free T4, and 24-hour radioactive iodine uptake establish the diagnosis. An elevated TSH concentration indicates primary hypothyroidism. Though relatively uncommon, hypothyroidism in the elderly may be secondary to pituitary tumor or pituitary-hypothalamic dysfunction. A low TSH concentration in the presence of low thyroid function studies should suggest this diagnosis.

The treatment of primary hypothyroidism is replacement with thyroid hormone. Currently, L-thyroxine is the most often used preparation. In younger individuals, the average maintenance dose is between 0.15 and 0.2 mg. once daily. Because of the risk of coronary insufficiency, myocardial infarction and cardiac arrhythmias, the initial dose of thyroid hormone should be considerably lower than the projected final maintenance dose. Gradual increases in dosage should be made at relatively long intervals (that is, three to four weeks). If adverse side effects should develop before the patient becomes euthyroid, the dosage should immediately be decreased to the next lowest dosage that did not produce adverse effects (37). In treating the elderly, it is frequently necessary to settle on a final maintenance dosage of thyroid hormone that is lower than the average maintenance levels in order to prevent cardiac compromise.

A rare but often fatal complication of myxedema is myxedema coma (41). It occurs more often in

women, in patients who have had longstanding, untreated myxedema and in older myxedematous patients. The mortality rate in patients with myxedema coma is reported to be between 50 and 70%. Once diagnosed, myxedema coma must be treated as a medical emergency. Treatment consists of either L-thyroxine given intravenously or L-triiodothyronine given either intravenously or via nasogastric tube. Recommended dosage schedules of L-triiodothyronine range from 10 ug every 12 hours to 100 ug every six hours. A reasonable schedule is 12.5 to 25 ug every six to eight hours. When using L-thyroxine, a reasonable schedule is an initial intravenous dose of 500 ug, followed four days later by institution of daily doses of 50 ug, intravenously, or 100 to 200 ug, orally. Needless to say, basic medical support and treatment for associated problems such as hyponatremia, hypoglycemia, ventilatory insufficiency, hypotension and shock must be given. Careful monitoring of cardiac status is essential. Patients in myxedema coma should be given 100 to 300 mg of intravenous hydrocortisone per day to counteract possible relative or absolute adrenal cortical insufficiency (37). Hypothermia in an elderly myxedematous patient frequently portends the onset of myxedema coma. This is an ominous sign. Temperatures as low as 74°F have been reported in elderly myxedematous patients. Interestingly, the patients with hypothermia secondary to hypothyroidism do not shiver. The hypothermia should be treated with blankets to prevent further heat loss. Active warming of the patient may be hazardous because peripheral vascular collapse and increased oxygen consumption may ensue without corresponding thyromimetic effect (42, 43).

Thyroiditis

As has been previously mentioned, Hashimoto's (lymphocytic) thyroiditis probably accounts for a sizable percentage of the hypothyroidism seen in the elderly. However, most cases of Hashimoto's thyroiditis do not originate beyond the sixth decade.

Subacute thyroiditis usually occurs in women in the third to fifth decades of life. It is usually characterized by tender thyromegaly, an outpouring of thyroid hormone into the periphery and essentially no uptake of radioactive iodine. A variety of this disorder which has been called "Silent thyroiditis" has also been described. A single case report of the "silent" variety has recently been reported in a 93-year-old woman (44). Overall, subacute thyroiditis is seen infrequently among the elderly. Riedel's

Struma or fibrous thyroiditis is a rare entity occurring in females two to four times more frequently than in males. The average age is approximately 50 years and cases have been reported into the eighth decade.

It is characterized by a painless, fixed, hard goiter which progressively enlarges. Clinically, it may be indistinguishable from thyroid carcinoma. The etiology is unknown. Thyroid function studies are usually normal but scan of the thyroid gland shows the involved areas to be "cold." The erythrocyte sedimentation rate is frequently elevated. Surgery is indicated to rule out carcinoma and to relieve tracheal compression. Corticosteroid and thyroid hormone therapy have been used with no apparent success (45).

Conclusion

As indicated, the thyroid disorders of the elderly do differ in many respects from their counterparts among younger individuals. It is of great importance to the clinician dealing with an elderly population of patients to keep in mind that thyroid disorders frequently present with manifestations more suggestive of disease entities of other organ systems. When there is palpable pathology of the thyroid gland, the physician will have no difficulty in following through appropriately. The areas in which thyroid disorders are more frequently overlooked are hyperthyroidism and hypothyroidism because of the subtle symptoms seen in the elderly. By remembering the key features outlined above, proper identification and management of these patients can be accomplished.

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Geriatric Psychiatry—an Overview

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One of the major developments in geriatrics during the past three decades has been the awakening of the scientific community and the public to the rapid growth of the geriatric portion of the population. Within medicine, frequent warnings of an epidemic of psychiatric illness in the aged have been sounded. As we begin a new decade, there are more than 1,000,000 patients over age 65 in long-term care institutions in the United States, over half of whom have psychiatric disorders. Kay (1) estimates that 10% of those over 60 will eventually suffer severe mental deterioration. The suicide rate is highest among aged white males. Delirium, a term in use a century ago, then renamed acute brain syndrome, is once again in use frequently as systemic illnesses and drugs used to treat them create confusion in significant numbers of the nondemented aged.

In response to such needs, a new interest in geriatrics has developed in psychiatry. As American psychiatry moved away from orthodox psychoanalytic theory, which held that anyone over 50 was too old to be suitable for psychoanalysis, more flexibility in technique developed and with it more optimism for psychotherapy of the old. Two members of the Psychiatry Department of Mount Sinai School of Medicine made important contributions to this change. M. Ralph Kaufman (2) found that transference did occur in older patients, along with fantasies of the therapist as their child.

Alvin I Goldfarb (3) used adaptional psychodynamics to build an entirely new framework for brief, supportive psychotherapy for the elderly. Most of his early clinical experiences were at the Jewish Home and Hospital for Aged, in New York City, where he established the first psychiatric department within a nonprofit long-term geriatric facility in the United States. He recognized the need to

integrate psychiatric diagnosis and recommendations with the ongoing care provided for patients by other physicians and by nurses, social workers and allied health professionals. The model of psychotherapy in office practice, psychiatrist and patient talking in complete isolation, was dropped because it precluded that integration. What evolved was an emphasis on team evaluation of problems presented by patients, families or institutional staff, then recommendation by the geriatric psychiatrist of changes in patient care. Such changes are carried out by those staff members best able to do so, in the judgement of the unit's team. Individual psychotherapy by the psychiatrist is recommended only for a small percentage of patients, both because of the scarcity of available psychiatric time in geriatric facilities and because it is not usually judged to be the most effective approach to institutionalized geriatric patients. The Jewish Home and Hospital for Aged program of psychiatric services has served as a model to many other geriatric facilities, both nonprofit and private.

A concurrent development has been multidisciplinary investigation into the interrelationship of mental and physical function, social factors and economic status in the aged. Contributions from medical research, sociologic and social work studies and economists have substantiated clinical observations that these factors all have vital influences upon the well-being of the aged and that such interactions affect mental function more strongly in the aged than in younger populations. While lost relatives and supportive friends cannot be replaced, the effect of such losses upon the self esteem and independence of the aged can be appreciated and understood. While lost physical function usually cannot be restored, the effect of impaired physical capacity upon the mood and motivation of the aged can be considered and acknowledged. The loss of working status and the anxiety of existing on a fixed income in inflationary times similarly needs to be understood in any holistic approach to the aged.

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These three areas are recognized as central to the understanding of the psychodynamics of aging. They produce, to a degree determined both by the patient's previous level of adaptation and by the severity of the losses caused by aging, a loss of mastery (coping ability), anxiety and depression. Frequently functional psychiatric disorders, especially depression and "problem behavior" result.

Within medicine, few still ascribe the major role in loss of physical function to the aging process, in sharp contrast to past eras. As diseases have become better understood, disease, not aging per se, has been recognized as limiting physical capacity at any age, even in the eighth and ninth decades. The more physicians have looked for evidence of disease in the aged, the more they have found. Active treatment of such diseases in the aged has, however, had mixed results. The multiplicity of disease common in the aged patient often leads to polypharmacy, drug interactions and failure to appreciate the altered pharmacology of the aged, with resultant intoxications with what would be well-tolerated dosages in younger patients. These plus inadvertent errors in self medication have resulted in hospitalizations for drug-induced problems in patients over 60 at a rate 50% greater than that for younger patients (6).

Psychiatry has participated in the shift to recognition of the aged. Therapeutic nihilism toward "senility" has undergone a marked change as active research has been undertaken. Katzman and Terry (7) have demonstrated that Alzheimer's disease is not limited to presenile patients, that in fact it is neuropathologically indistinguishable from the brains of older demented patients. With this, investigations into the pathophysiology of what is now widely referred to as senile dementia, Alzheimer's types are in progress.

At the same time, efforts to understand the basic neurophysiology of memory have produced new findings. Most research has been done on three groups of substances: nootropics, neuropeptides and neurotransmitters. Of the first group, piracetam (2-oxopyrrolidone acetamide) has been shown to have a specific positive effect on learning in healthy young subjects (9). It may promote brain energy reserves by increasing the ratio of ATP to ADP. Piracetem has been used in a variety of clinical studies with virtual absence of any known toxic effects, in any dosage. Studies of its effect on "normal" aging subjects and on patients with mild to moderate cognitive decline have been encouraging but not conclusive; studies on severely impaired patients have shown no effect (10).

Of the neuropeptides, an ACTH moiety (ACTH 4-10) devoid of peripheral hormonal effects has been under active investigation. Despite animal studies showing positive effects upon both memory and learning, and electrophysiologic data indicative of CNS effects, thus far no useful effects in the cognitively impaired aged human have been demonstrated. Other animal studies have suggested a role for vasopressin in memory. It has been administered as a nasal spray to patients with posttraumatic amnesia with dramatic benefit (11). Preliminary work in elderly humans indicates vasopressin does improve memory but a controlled study has not yet been reported (12). The enkephalins, too, appear to have a role in memory. Their anti-amnesic effect is not blocked by Naloxone.

Some evidence implicates each of the four known neurotransmitter systems of the brain in age-associated cognitive decline, if not in senile dementia, Alzheimer's type. Dopaminergic deficiency is a major etiologic factor in the Parkinsonian syndrome. Drachman has found improvement in cognitive function in senile demented patients having Parkinsonian symptoms, treated with L-Dopa (13). However, it is the cholinergic system that has attracted the most study. Scopolamine, an anticholinergic agent has long been known to produce amnesia. Physostigmine has been shown to reverse anticholinergic-induced confusion. Davis and Mohs, now working at Mount Sinai, have shown that dosage is critical in its effect on memory (14). They and others are currently conducting double-blind controlled studies to evaluate the effects of specific neurotransmitter agonists on the cognitively impaired aged.

As in most new areas of research, these initial findings have raised more questions than they have answered but they have already made it clear that psychophysiology is providing new leads toward a psychopharmacology of cognitive impairment, with sound reason for optimism for effective treatment of such disorders in the aged in the foreseeable future.

With exception of cognitive impairment, effective treatment for psychiatric disorders of the aged already exists. Treatment of depressive reactions is as effective in the aged as in younger patients, provided attention is given to the special psychologic and metabolic differences accompanying aging. The former have been touched on earlier in this article. Decrease in hepatic detoxification and in renal excretion rates reduce the ability of most aged patients to tolerate standard dosages of antidepressant drugs, especially the cyclics. Since tricyclics can produce or accentuate cardiac arrhyth-

mias, the aged depressed patient's cardiac status should be carefully monitored when they are used. Doxepin is probably the safest antidepressant for the aged, especially when initiated in small doses. Those who do not respond to tricyclics may be given MAO inhibitors if closely followed. Those who fail with all antidepressants merit electroshock treatment. It is also the fastest, most effective treatment for severe agitated depression. Unipolar EST, rarely needed more than five to six times for such patients, does not produce lasting additional memory dysfunction, even in the presence of pre-existing cognitive impairment. Lithium is indicated for manic-depressive patients, some of whom continue to have clinical episodes in extreme old age.

Tardive dyskinesia is more frequent among older patients. Phenothiazines should not be used to control agitated behavior or even psychotic symptoms in aged patients unless strenuous efforts have been made to change them by other means, such as environmental and attitudinal changes, and the use of minor tranquilizers. If a phenothiazine is prescribed, the dose should be as low as possible. It should be reduced and then discontinued as soon as practicable. Thiothixene appears less prone to produce tardive dyskinesia. In those patients in whom it does occur, choline may be beneficial.

While not marked by such obvious side effects, the use of benzodiazepines in the aged is not innocuous, unless carefully administered. Long-term use should be questioned, particularly if it is requested not by the patient but by relatives or by the staff of a geriatric facility. Inquiry often leads to the discovery of personality problems better dealt with in ways other than tranquilizing the aged patient.

The advances in geriatric psychiatry described above have led to new interest in training programs for geriatric psychiatry within psychiatric residencies. Finkel (15) has described such a program at the Michael Reese Hospital in Chicago. In listing the content to be taught, he summarizes the complex, yet vital considerations those working in geriatric psychiatry must master: Developmental Goals of Later Life, Theories of Aging, Social and Psychological Effects of Perceptual Changes and of Chronic Physical Illness, Family Evaluation, Role Reversal, The Impact of Retirement, Of Widowhood, Attitudes toward Death. Blumenthal (16) stresses the need for educating new geriatric psychiatrists to work closely with internists, family

physicians, nurses and social workers and to gain knowledge of community resources for the aged. While experience in a general hospital can provide a base to reach some of these goals, it does not afford experience in the problems of caring for long-term patients. Only clinical training in skilled nursing facilities will enable psychiatric residents to understand the special needs of the large numbers of the aging population who are institutionalized. Similarly, medical students who do not spend some time in skilled nursing facilities miss an important aspect of education in geriatrics.

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Some Observations on Cardiology in the Aged

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The human life span has not increased significantly for centuries. What has changed is the average life expectancy, thus allowing more people to reach the limit of what appears to be a fixed life span with a maximum somewhere near 90 or 100 years. Although death may result from cardiovascular disease, this condition arises largely from the age-related degeneration of arterial walls. The fundamental cause of death in the later years is attributed to the body's increased vulnerability to disease resulting from the inexorable decline in its functional capacity. The likelihood of dying doubles every eight years after the age of thirty. The defense of the body's increased vulnerability to disease, with advancing age, has been especially vigorous in the field of cardiology. The more successful this defense, the greater will be the impact on governmental agencies that currently spend 132 billion dollars on a constituency of 24 million people over 65 years of age, who comprise only 11% of the population in the United States today. If present policies continue at the same level, annual federal expenditures on the elderly will triple by the end of the century and would then represent 40% of total federal spending.

The Recent Decline in Cardiovascular Mortality

Beginning in the 1940s, coronary heart disease rose relentlessly in the United States. By 1960, arteriosclerotic heart disease, the leading cause of death in the United States, accounted for 29% of the total number of deaths for that year, 30% of which occurred before the age of 65 years. In recent years, a dramatic reversal of this trend has taken place, beginning in the mid-sixties (1, 2). The fall has been most precipitous since 1968 when mortal-

ity from the major component of cardiovascular mortality, coronary artery disease, began to decline. Age-adjusted death rates for coronary artery disease in the United States have been lower each year for nine consecutive years, and the decline involves all age groups. The implications of this statistical trend are quite obvious for those interested in cardiology in the aged.

Death rates from rheumatic heart disease began falling early in this century, long before penicillin prophylaxis was in use. A steady decline in the mortality rate from hypertensive heart disease and its major complication, stroke, began prior to the introduction of effective antihypertensive medications. These long-standing trends suggest environmental factors rather than specific medical interventions to account for the improvement. Cardiovascular mortality varies widely throughout the world (the rate is actually increasing in many European and Scandinavian countries) suggesting that the wide spectrum of diseases of the heart is not an inevitable consequence of aging. Migrants from low-incidence areas are found to assume the higher incidence of their newly adopted countries.

Major factors related to the declining coronary mortality rate include coronary care units, antihypertensive programs, decline in cigarette usage, dietary changes and improved diagnosis and therapeutic modalities. However, the data supporting the contribution of coronary care units to significant lowering of coronary death rates is weak, (3) particularly in the aged (4). In the Framingham study, comparing periods before and after the introduction of coronary care units, overall coronary heart disease mortality fell from 8.4 per 1000 at risk to 7.7, when figures were age-adjusted for men 50-74 years of age. The case fatality reduction was also not statistically significant (31% falling to 26%).

The level of risk factors in the United States

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population seems to be falling. Older people appear to be taking more exercise; obesity is less common; animal fat in the diet has been reduced paralleling a reduction in overall serum cholesterol; smoking is less popular; blood pressure reduction programs are widespread. The elderly share the declining cardiovascular mortality with the overall population of the United States, strongly suggesting that risk factor modification even in advanced age may be effective, or that better medical care is responsible for reducing mortality once an attack occurs. It is important to emphasize that what is declining is mortality (1). At present, there is no conclusive data on the incidence of new coronary events. A decline in incidence rate would imply that the improvement is caused by primary prevention (lifestyle change, antihypertensive therapy). On the other hand, if the incidence rate is steady, or rising, then the decline in mortality rate has to be from improved medical care.

Although numerous reports have been presented relating risk factors to cardiovascular death rates, remarkably little is known of the inverse relationship: have people who lived unusually long lives in apparently good health shown few coronary risk factors during life? A study of healthy, elderly people, 90 years old or older by Danner et al (5) revealed a very low cholesterol and an elevated high density lipoprotein:total cholesterol ratio. Whether this lipid pattern represents selection or is the result of aging itself is not clear.

Systolic Hypertension in the Aged

More than 50% of the patients with cardiovascular disease have hypertension and even more have underlying atherosclerosis. About 20% of American adults have arterial hypertension and there is considerable evidence that the control of blood pressure will prevent many of the cardiovascular complications of this condition. It has not, however, been established that antihypertensive therapy reduces the atherosclerotic complications of hypertension (6).

Hypertensive vascular disease and atherosclerotic vascular disease are separate entities and cause distinguishable pathologic changes in the arterial tree. One major difference is the accumulation of lipid in atherosclerotic lesions and the absence of excess lipid in purely hypertensive lesions (6). Hypertension accelerates the progression of atherosclerosis and the atherosclerotic complications (coronary disease, myocardial infarction and stroke) are more difficult to control than the hypertensive complications (left ventricular hypertrophy, congestive

heart failure, dissection, intracerebral hematoma, renal failure).

Hypertension associated with arteriosclerotic vascular disease is a major cause of cardiovascular morbidity in older persons. It appears to be one of the major complications of aging and results from inelastic large arteries, and the increased rigidity of the larger vessels leads to an elevated systolic blood pressure, with a markedly widened pulse pressure. At any level of diastolic pressure, the risk for cardiovascular mortality rises in direct proportion to the systolic pressure (7). Sarnoff (8) indicated that increased systolic pressure generation and developed tension was much more demanding in terms of energy requirement than is increased diastolic filling pressure. Evidence associating pure systolic hypertension with increased cardiovascular mortality is irrefutable, but there is no proof of a cause and effect relationship.

In the isolated systolic hypertension of the aged, there is room for a legitimate difference of opinion regarding the merits or value of antihypertensive treatment. Drug treatment is likely to be less successful in reducing the elevated systolic pressure levels in patients with a fixed, structural basis for their arterial rigidity, and aggressive treatment may lead to reduced cardiac output or a further widening of the pulse pressure, by primarily reducing the diastolic blood pressure. Although the increased morbidity and mortality associated with systolic hypertension has been adequately demonstrated, no long-term controlled study has been carried out to assess the benefits of therapy of systolic hypertension, primarily associated with rigid vessels. Aggressive therapy leading to excessive reduction of blood pressure may precipitate vertigo, orthostatic hypotension and on occasion a cerebrovascular accident. It is not uncommon for elderly patients to complain of marked accentuation of symptoms due to cerebrovascular insufficiency, even on moderate doses of antihypertensive medication.

Not uncommonly, the diagnosis of hypertrophic cardiomyopathy is missed in an elderly hypertensive, who has a systolic murmur and syncopal attacks. This condition is compatible with long life (or the obstructive process develops progressively late in life) as shown by several series of patients in their seventh and eighth decades (9, 10). The condition is frequently misdiagnosed as calcific aortic stenosis, hypertension, angina pectoris or mitral insufficiency. The diagnosis is made by the characteristic echocardiogram demonstrating asymmetric septal hypertrophy or systolic anterior motion of the mitral valve and propranolol therapy often leads to symptomatic improvement.

Pathology of the Aged Heart

In addition to the standard and well recognized pathologic entities of ischemic, hypertensive and rheumatic heart disease and endocarditis of infectious origin, certain more-or-less unique conditions are encountered in the elderly patient. These include massive calcification of the mitral annulus, senile cardiac amyloidosis, thrombotic nonbacterial endocarditis and sclerosis of the heart valves of indeterminate etiology (11). In addition, cardiac rupture is highly age related.

A distinction should be drawn between the pathologic entity of degenerative valve sclerosis, and calcification of the valve with distortion and commissural fusion. In the former, quite common condition involving the mitral and aortic valves in the elderly, leading to loud systolic murmurs, at apex and base, but without significant hemodynamic effects, deposition of the calcification in the cusp fibrosa begins centrally. Varying degrees of rigidity and immobilization of the cusps occur without reduction in orifice area. On the other hand, in calcific aortic stenosis, one finds heavy calcification superimposed on commissural fusion leading to marked infringement on the aortic orifice area. Roberts (12) believes that many of these cases originate as congenitally bicuspid valves which calcify with advancing age. This is especially true if the aortic involvement is anatomically isolated, and is not associated with mitral valve involvement.

Senile cardiac amyloidosis appears to be a clinically distinct variety of primary amyloidosis occurring in patients of advanced age with the amyloid almost exclusively limited to the heart, compressing and replacing myocardial fibres. Pomerance (13) found it present in 18% of the autopsied elderly patients with heart failure and in 5% of those without. The incidence rose with age and occurred in 71% of a group of 14 patients over 90 years of age with heart failure. Five percent of patients with cardiac amyloidosis have been reported to have severe occlusive amyloid deposits in intramyocardial arteries leading to localized areas of ischemic necrosis and intractable heart failure. A recent report indicates that a virtually diagnostic feature of amyloid heart disease in older patients with unexplained heart failure is a "granular sparkling" appearance of thickened ventricular walls and septum on two-dimensional echocardiography (14).

Calcification of the mitral annulus leads to varying degrees of mitral insufficiency. The mechanism appears to be displacement of the posterior leaflet of the mitral valve into the left atrium and interference with proper mitral ring contraction. Conduc-

tion abnormalities are frequent since the junction of the mitral ring and the membranous interventricular septum become involved early in the calcification process, and heart block is not unusual. In a group of 31 patients over 60 years of age with mitral annular calcification 51% demonstrated conduction defects (15). Patients, particularly of advanced age, with radiologic and echocardiographic evidence of calcification of the mitral annulus may be at particular risk of cerebral or retinal ischemia, probably because of multiple systemic emboli arising from extruded material from the massively calcified mitral ring (16). A recent cardiac ultrasound study indicates that in most cases, the bulk of the calcific deposit is located not at the fibrous mitral ring but rather in the angular space between the posterior mitral leaflet and the subjacent left ventricular posterior wall (17). These workers suggest that the term submitral calcification be substituted for mitral annulus calcification.

Free transmural rupture of the heart is reported to occur in as high as 24% of fatal acute myocardial infarctions. After fatal arrhythmia and cardiogenic shock, it is the most frequent cause of sudden death in acute myocardial infarction, particularly in the aged. Rupture of the free wall of the heart leading to sudden hemopericardium is eight to ten times more common than rupture of a papillary muscle or the ventricular septum (18).

While estimates in the literature vary, the majority of cardiac ruptures take place about the fifth day following an acute infarction (19). More than 90% of these penetrate the free wall of the left ventricle. The pathway of rupture is often circuitous and hemorrhagic, leading some authors to speculate that the rupture may be the result of a dissecting intramural hematoma. Recent coronary arterial occlusion leading to transmural infarction in a left ventricle with poor collateral circulation and a notable absence of fibrosis in the infarct seems to be the most frequent substrate leading to rupture. Schuster and Bulkley (20) suggest that infarct expansion predates rupture by several days and can be detected clinically before rupture takes place by two-dimensional echocardiography. A surprisingly high incidence of rupture has been found in autopsies on elderly, institutionalized, psychotic patients, suggesting that continued physical activity after clinically undetected infarction may predispose to rupture.

Age is a significant unfavorable risk factor for myocardial rupture. It is seldom seen before the sixth decade and its peak incidence occurs during and after the seventh decade of life (21) and has a greater frequency in women than men. Hyperten-

sion during the acute phase of infarction and the absence of significant evidence of congestive heart failure were noted. The classical clinical picture of the patient suffering a free wall rupture of the heart is characteristically that of an older female who sustains her first uncomplicated myocardial infarction and suddenly develops renewed severe chest pain on the 4th or 5th day, associated with hypertension, and conduction abnormality. The picture of cardiac tamponade rapidly develops due to hemopericardium. Prompt echocardiography, pericardiocentesis and emergency cardiac surgery would appear to be the only hope, albeit a heroic one, of increasing survival in this dread complication of myocardial infarction.

Pacemakers

The current indications for cardiac pacemakers have expanded recently as improvements in technology, miniaturization, advanced electrode design and improvement in circuitry have broadened pacemaker application. In patients with complete heart block, particularly if Adam-Stokes syndrome or congestive heart failure are present, there appears to be little disagreement that a pacemaker should be implanted. The sick-sinus syndrome, characterized by symptomatic sinus bradycardia, sinus arrest or the brady-tachycardia syndrome accounts for almost one-third of the present pacemaker implantations. Fascicular blocks that develop during acute myocardial infarction or when accompanied by syncope are another indication, and to this can be added Mobitz 2 phenomenon. Recently, patient-controlled radio-frequency pacemakers with a rate maximum of 400 impulses a minute have been used to interrupt recurrent paroxysmal atrial tachycardia. Pacemakers have been implanted as a back-up to the administration of drugs in the treatment of patients with severe angina pectoris who cannot tolerate propranolol because of excessive bradycardia with this agent. Similarly, elderly patients that require digitalis to control rapid supraventricular rhythms at times may be protected from symptomatic bradycardia, with a pacemaker.

It is physiologically and hemodynamically rational to choose atrial pacing in patients with symptomatic bradycardia, if there is no evidence of A-V block. In patients with sinus bradycardia, the cardiac output can only be augmented by atrial pacing, because of the atrial contribution to ventricular filling. Unfortunately, many older patients subsequently develop A-V block even if there is no suspicion of such block at the time of implantation of

the atrial pacemaker. A-V sequential pacemakers are the obvious solution in such patients, but multiple electrodes are required and tip displacement from the atrium is common.

An infrequent, but important, complication of ventricular pacing in the older patient with sick sinus syndrome is a dramatic and life-threatening fall in cardiac output and arterial pressure following pacemaker insertion (22). We have seen such a patient who developed severe arterial hypotension in the recovery room following the implantation of a ventricular pacemaker, leading to convulsions. With the substitution of atrial pacing, from the coronary sinus position, normal output and arterial pressure was promptly restored. This paradoxical result of ventricular pacing may be related to the loss of the atrial contribution to cardiac output, or to an atrial reflex initiated by atrial contraction against closed atrioventricular valves leading to a sharp fall in peripheral resistance.

The feasibility of home-checking by the patient of his own pacemaker has become an imperative with the large number of pacemaker patients. Monitoring of pacemaker intrinsic rate to determine battery dysfunction can easily be accomplished with a transistor radio and a magnet. There are also a large number of devices that can transmit signals to the physicians office to provide "instant electrocardiograms" for analysis of pacemaker function.

It is of interest to note that pacer patients have life expectancies at least equal to those achieved by non-pacer individuals, of matched age. Some statistics even suggest that the survival of the pacemaker patient may even be longer than his unpaced control (23). The change from a mercury-zinc to a lithium power source has extended the average life of pacemaker generators to over six years and the failure rate has become quite low. Median survival was 5 years in patients older than 74 years when their pacemaker was implanted (24). Nuclear powered pacemakers are not practical for the patient of advanced age since they are devised to last 30-40 years.

Electrodes—the weakest link in the pacemaker chain of events because of breakage and perforation—are being improved. A three-year study by three medical centers has revealed a one-year electrode malfunction rate of 7.4% (25). Early electrode malfunction may be infrequent, or it may occur in as many as 56% of cases. A 10% rate is typical. Direct epicardial pacemaker implantation has a higher risk of mortality and morbidity than transvenous positioning, but may have to be resorted to particularly in a patient with a large right ventricle

and congestive heart failure. More improvement in electrode technology is required. The damage caused to the myocardium by the constant pressure applied by a stiff or a flanged lead, results in fibrosis contributing to a rising chronic threshold. Fracture of such leads was as high as 20% in ten years. Smaller leads, easier to pass through small veins, and leads that can be used in multiples in one vein for A-V sequential pacing are needed. Endocardial screw fixation with a high flex-life lead is being developed. Tined atrial leads unfortunately require a large calibre vein, such as the external jugular. These are required to keep the tip in place in the atrium for reliable pacing. The same problem exists for A-V sequential pacing leads.

The Electrocardiogram in the Aged

Mihalick and Fisch (26) found that the incidence of clinical heart disease and an abnormal electrocardiogram increased with age in a parallel fashion. They found significant correlation with left bundle branch block, nonspecific conduction disturbances, S-T and T wave changes and atrial fibrillation. There was no significant correlation with left anterior hemiblock, right bundle branch block, first degree A-V block, or frequent premature contractions. Golden and Golden (27) found a history compatible with myocardial infarction in only 11% of subjects over the age of 90, with definite electrocardiographic changes indicating this diagnosis.

Perloff et al (28) in assessing left axis deviation found a gradual leftward drift of the QRS, as age increases, governed primarily by conduction through the left anterior fascicle. During aging, conduction slows through this fascicle, with or without left ventricular hypertrophy. Pure concentric hypertrophy of the left ventricle causes increased QRS voltage, but not a leftward shift in the frontal plane. Many pathologic states can also cause left axis deviation, but all such effects on the left axis shift must be interpreted with age in mind.

Cardiac Drugs in the Aged

The aged appear to be more sensitive to many drugs than the young, and this is particularly true of digitalis. The marked increase in the prevalence of cardiovascular pathology in aged man requires extensive use of cardiac glycosides in this group. Studies on glycoside activity appear to indicate altered sensitivity of the myocardium to the effects of digitalis preparations with advancing age. Serum levels of the glycosides, for a given dose, have been shown to increase in advanced age (29).

Guarnieri et al (30) demonstrated a diminished inotropic responsiveness to intravenous acetylstrophanthidin in senescent dogs, but no difference in threshold for toxicity to this digitalis preparation. In the aged, therefore, the same risk of toxicity for a given serum level exists, but the inotropic benefit from digitalis is reduced. In view of the uncertain bio-availability of digitalis preparations in the aged because of altered gastrointestinal absorption, renal clearance or volume distribution, attempts to achieve maximum therapeutic effects with digitalis are risky in view of the decreased inotropic-toxic ratio in the elderly cardiac.

It has also been shown that reduced clearance of quinidine and prolongation of its elimination half-life could easily predispose to toxicity in the elderly unless the dose is carefully titrated (31). The combination of quinidine and digoxin seems to be particularly hazardous (32) as shown in studies suggesting that tissue-bound stores of digoxin are liberated into the serum and can quickly rise to toxic levels in old patients given quinidine and digoxin simultaneously. Some exciting new work on the reversal of digitalis toxicity by specific antibody fragments may be of particular value in the aged (33).

It is general practice for clinicians to continue the administration of digitalis indefinitely once it has been started. This practice has been put to test recently in groups of elderly patients who have been on chronic maintenance digoxin therapy for many years. The discontinuance of digoxin in elderly patients who had been on the drug for many years was tolerated in a large percentage of patients, particularly those in sinus rhythm (34). However, in patients with established atrial fibrillation, grossly enlarged hearts or with recurrent supraventricular arrhythmias, digoxin could not be terminated with impunity. Management of congestive failure is surprisingly effective, in many elderly patients in sinus rhythm, with the use of diuretics alone.

Cardiac Surgery in the Aged

With the introduction of potassium-enriched ice-cold solution to irrigate the coronary arteries, a significant reduction in mortality took place not only in the general cardiac surgery population, but more particularly in the aged. Numerous reports indicated that the mortality of patients 65 years and older following valve replacement began to approach that of the younger patients. The addition of coronary by-pass grafting to the valve surgery

however caused a significant rise in mortality rate and the combined approach appears to be risky (35) although this appears to be considerably reduced in subsequent series utilizing hypothermic cardioplegia.

Experience varies and in one large series of 89 elderly patients (mean age 72.8 yr.), mitral valve replacement carried an unusually high mortality of 56%, which was seven times greater than patients less than 70 (36).

Surgery in any aged group is technically more difficult for many reasons related to anaesthetic administration. Aged patients quickly become hypothermic if chilled in the operating room. They are more sensitive to narcotic drugs and anaesthetic agents. Absorption of oral agents is unpredictable because of reduction of gastric, pancreatic and oropharyngeal secretions. Postoperative hypoxemia tends to increase with advancing age. Old age alone has been declared an indication for endotracheal intubation, and assisted ventilation in the postoperative period is of extreme importance. Postoperative acidosis is frequently associated with poor renal function. One-third of surgical patients over 70 years of age have some disturbance of cardiac rhythm. The postoperative complication rate in patients with previous myocardial infarction is high (37). Postoperative deterioration of neurologic status is disturbingly high in the aged. This may be related to hypotensive episodes during surgery and must be guarded against.

An important consideration in recommending cardiac surgery in the very aged, is that it appears some disease processes tend to progress more slowly in the elderly, a fact previously recognized in some forms of cancer. For instance, with abdominal aortic aneurysms, the time from diagnosis to rupture in patients over 65 years of age who refuse surgery is twice that of patients under 65 (30 months vs. 18 months). Paradoxically, the patency of arterial bypass grafts is maintained at a higher rate over the years, in the elderly (38). The results of emergency cardiac surgery in the aged, however, are bleak (39).

Is There Something Unique about Heart Disease in the Aged?

Burch (40) has stated that the major difference between heart disease in the aged and the young is that the elderly generally have one or more other major, potentially life-threatening conditions complicating the cardiac condition. These associated problems may mask or alter the presentation of classical syndromes in a way that may make them

difficult to recognize. Acute myocardial infarction is an example of this in that the diagnosis may be established beyond a doubt on other evidence, yet the elderly patient has complained of no pain whatsoever. It is not clear whether this is due to anatomical factors, or that mental confusion masks the symptomatology or whether there is a psychological rejection of the unpleasant experience. The fact remains that myocardial infarction is frequently silent in older people, when silence implies absence of cardiac pain.

This point is emphasized by Pathy (41) in a review of myocardial infarction in 387 elderly patients, in which only 75 had a classical onset with substernal, or epigastric pain. In the rest, the "silent coronary" appeared with a variety of symptom complexes including sudden onset of mental confusion or coma, pure dyspnea without pain, sudden hemiparesis, vertigo, recurrent vomiting, renal failure, intense weakness, restlessness or profuse sweating. The sudden appearance of gangrene of an extremity, or mesenteric occlusion, should alert the geriatrician to the possibility of an embolus arising in the mural thrombus occurring in acute myocardial infarction.

Harris (42) suggests that although no single abnormality characterizes the aging heart, cellular, physiologic and anatomic changes support the hypothesis that senile cardiopathy exists. Elasticity of the aortic compression system is only half as great at age 70 as it is at 20. Heart valves thicken and become more rigid with advanced age. Atrophy of the myocardial cells occurs and this may reduce the compliance of the ventricular cavities and a delayed recovery of contractile ability. A decrease in the amount of muscle and a corresponding increase in fibrous tissue in the sinoatrial node and conduction system has been noted in aged hearts. Atrophic changes in the mitochondria, a decline in enzymes required to support contractile properties, and less efficient mobilization of catecholamines necessary to augment the speed and force of contraction under stress are noted in the senescent heart. Taken together, one can construct a strong case for the cardiopathy of aging.

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Gastroenterology in the Aged

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Aging produces general and specific effects on the morphology and physiology of the gastrointestinal tract, its symptoms, and responses to specific diseases. The incidence and management of these effects are also profoundly influenced by the aging process. Selected examples of these factors will be used to help the clinician better understand and manage them as they occur in the geriatric patient. General surgery, including gastrointestinal, and chemotherapy of malignancies will be reviewed in other sections of this issue and will not be considered here. The chapters on gastroenterology in the increasingly available books on geriatrics (1-3), as well as the comprehensive and specialized texts on gastroenterology may be used for more detailed or fuller reviews of the topics covered here as well as some that are not.

With aging, there is a variable decrease in the secretion of digestive enzymes by the stomach, intestines, pancreas, and liver and that of acid by the stomach. This may result in diminished absorption of some nutrients. Mucosal atrophy and decreased blood supply may also interfere with absorption. Decreased motility of the hollow viscera, in addition, may produce a variety of symptoms. Most gastrointestinal symptoms increase with aging. Some of this increase is caused by the altered physiology, but the increased prevalence of specific diseases such as cancer, hiatus hernia, and colonic diverticular disease also contribute to the increased symptomatology.

Symptoms referable to the digestive tract become increasingly bothersome to aging patients and may emotionally disturb them. Although functional disorders are the most common cause of gastrointestinal symptoms in the aged (4), persistent symptoms should always be investigated. Many of them

will be caused by organic disease. Even if they are found to be functional, an attempt should be made to treat those symptoms, rather than dismissing them as being of no significance and part of the "normal aging process." With meticulous attention to details, it is usually possible to afford significant symptomatic relief safely. Concomitant cancerphobia, whether overt or not, may also be relieved.

Compliance is necessary to obtain maximum benefit from therapy, whether directed toward a specific disease process or intended for symptomatic relief. Adequate compliance is frequently a problem in elderly patients with gastrointestinal disorders, just as it is in patients with other disorders. Similar techniques are useful to produce improvements. Precise and detailed instructions should be given in language readily understood by the patient. Specific details may be supplemented by written instructions. An understandable explanation of the problem and the rationale for the proposed treatment is also helpful. Therapy has to be devised in a manner that is practical for the patient to comply with.

Polypharmacy is also a problem in the geriatric population with gastrointestinal disorders. Aging changes the pharmacokinetics of many drugs, increasing the risk of adverse reactions (5). A comprehensive list must be made of all medications the patient is taking, being aware of the various possible side effects and interactions. This list should be reviewed periodically. Smaller doses and frequent monitoring are often needed to avoid drug toxicity in elderly patients. It is often discovered that the patient's heartburn is associated with the aspirin taken as a sedative and not considered as medication because it is nonprescription. A narcotic analgesic taken for dental pain may turn out to be the cause of the recently developed constipation, and an antihypertensive may be the cause of the patient's diarrhea. The often forgotten anticholinergic effect of tricyclic antidepressants may make an otherwise appropriate dose of an antispasmodic

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excessive. In addition, elderly patients with their various illnesses, who have been cared for by several physicians, may still be taking medications no longer necessary or even contraindicated for their current conditions. The cost of drugs must also be a consideration in prescribing for some elderly patients who may be unwilling or unable to pay for them because of financial difficulties.

Many of the recent technological advances in gastroenterologic diagnosis and therapy are of special value in geriatrics. Fiberoptic diagnostic gastrointestinal endoscopy has been found to be safe and effective in diagnosing lesions and discovering pathology in older patients and is often used as a complementary examination to standard barium radiology (6). Colonoscopy permits examination of the colon in those unable to retain sufficient barium for a satisfactory barium enema as well as permitting biopsy of questionable lesions already found. Extensive diverticulosis of the colon, which is much more common with increasing age, poses a particular problem in the patient with bleeding. Ruling out a concomittant small carcinoma or bleeding polyp is best done by colonoscopy. Fiberoptic upper gastrointestinal endoscopy has gradually become the investigation of choice in diagnosing the cause of upper gastrointestinal tract bleeding. The ability to biopsy with these instruments has greatly enhanced their use in establishing or refuting the presence of malignant disease.

Ultrasonography is becoming increasingly more valuable in the diagnosis of diseases of the pancreas and hepatobiliary tree (7). It is noninvasive, rapid, economical and has a high degree of accuracy in detecting gallstones and biliary dilatation. It is particularly useful in patients with jaundice, that precludes the use of standard oral cholecystography or intravenous cholangiography. The normal pancreatic duct can now commonly be imaged by ultrasound so that small lesions that produce obstruction and dilatation can be identified. It is especially valuable in recognizing pancreatic pseudocysts. Computerized axial tomography (CAT) scanning, another noninvasive technique, is being evaluated for demonstrating hepatobiliary and pancreatic pathology and is often complementary to ultrasonography. Its high cost and relative unavailability makes it of much less general usefulness at this time. Noninvasive nuclear scanning is particularly valuable for imaging of the liver and delineation of many of its lesions and, here, ultrasonography is often complementary.

Endoscopic retrograde cholangiopancreatography (ERCP) and fine-needle (Chiba) percuta-

neous transhepatic cholangiography are two invasive techniques that have become increasingly valuable in the diagnosis of hepatobiliary and pancreatic diseases and have been demonstrated to be safe in the elderly patient. They are reserved for situations in which the older standard studies and the newer noninvasive ones do not resolve diagnostic dilemmas. Arteriography is an invasive study that has less applicability in aged patients because severe atherosclerosis often makes the introduction of a catheter more hazardous or impossible. Ultrasonically or radiographically guided needle biopsies of pancreatic lesions can often establish a tissue diagnosis and avoid a diagnostic laparotomy.

Therapeutic polypectomies using fiberoptic endoscopes are probably of more overall value in younger patients, in whom the long term development of malignancy is more of a consideration. However, the safety of the procedure is such that bleeding lesions and suspicious ones can now be removed without undue risk to the older patient and abdominal surgery can be avoided. Percutaneous transhepatic bile duct catheterization in patients with obstructive jaundice caused by inoperable pancreatic carcinoma can be used as an alternative to surgery (8). Palliative long-term drainage can be accomplished either externally or internally through a prosthesis. Endoscopic papillotomy has become increasingly available as a safe and efficient means of dealing with choledocholithiasis, particularly in patients who are postcholecystectomy.

Diet and Nutrition

Whenever the diet and nutritional status of the elderly is considered, their lifelong eating pattern must be considered as well as changes that have occurred with aging. Although some age-related decreases in gastrointestinal absorption and function occur, they are usually small in comparison with the reserve capacity of the system (9). Poor diets in the elderly are usually associated with lower incomes, living alone, poor dentition, inactivity, and diets prescribed for diseases more prevalent in this age group such as diabetes mellitus, hypertension, and congestive heart failure. If poor nutrition is found, in spite of an adequate dietary intake, it must not be dismissed as a normal consequence of aging. An underlying disease process resulting in malabsorption must be sought because effective treatment is usually available. Unfortunately, careful research aimed at determining the requirements of most nutrients specifically for the elderly is just beginning (10). It is well known that, with increasing

age, gastric secretion of acid and of intrinsic factor decreases, associated with some degree of mucosal atrophy. Their association with iron and vitamin B12 absorption would indicate that recommended daily allowances of these nutrients should increase in the geriatric population.

Medical understanding of diets has changed in recent years, especially for diseases of the gastrointestinal tract. The classical bland diet is no longer prescribed for peptic ulcer disease. A high residue diet, rather than a low residue one, is now the diet of choice for diverticular disease. It is frequently difficult to convince a patient with one of these illnesses of many years duration to change from their previously prescribed diet to the currently recommended one. Careful explanations and a gradual transition is often the most effective course to follow.

Dietary fiber has become a topic of increasing interest in recent years. Some diseases of the gastrointestinal tract such as diverticulitis, cholelithiasis, and carcinoma of the colon are more common in the elderly and seem to be related to the amount of fiber in the diet, as one of many factors in helping to prevent these diseases. It should also help relieve the constipation that is so common in this age group. One or two tablespoons of bran a day is an effective way of increasing fiber in the diet.

A careful dietary history is, therefore, always important in elderly patients. If changes are recommended in their diet, they must take into consideration socio-economic, cultural, and ethnic factors as well as the purely medical ones. Some compromises may have to be made in order to find a diet that is practical as well as acceptable to the patient. Then, a rational discussion can be made as to whether vitamin and nutrient supplements will be necessary. Periodic reevaluations should be done to ensure compliance and to determine whether changes are required.

Esophagus

A number of esophageal diseases such as carcinoma, diverticula, diffuse spasm, and lower esophageal (Schatzki) rings increase in frequency in the elderly. Dysphagia is a frequent symptom of esophageal disease and must always be investigated, since it is almost invariably associated with organic disease. However, in older patients it may be caused by extraesophageal disease such as pseudobulbar palsy due to cerebrovascular disease. Diagnosis and therapy are essentially the same as in younger

age groups, although associated or concomitant disease may preclude surgical intervention. Esophageal motility is often markedly diminished in the aged patient. However, much of the decrease may well be from associated diseases that are more common in the elderly—such as diabetes mellitus or neurologic disorders rather than directly from the aging process (11). Differentiation between cardiac and esophageal chest pain is occasionally difficult. An esophageal acid perfusion (Bernstein) test may be used for this differentiation if standard symptomatic treatment does not elucidate the problem. However, it must be remembered that nitroglycerine can sometimes relieve the pain of esophageal spasm.

The incidence of hiatus hernia increases with increasing age, and may be detected in up to 70 percent of people over age sixty five. It is now accepted that many hiatus hernias are not associated with significant symptomatology. When present, the main symptoms are those of gastroesophageal reflux, mainly heartburn and acid regurgitation. Although most patients with reflux have a hiatus hernia, reflux can occur without a demonstrable hernia and the majority of patients with hernia do not have reflux. Reflux esophagitis is believed to result from insufficient pressure of the lower esophageal sphincter allowing acid reflux. In recent years, Urecholine, which increases the tone of the lower sphincter, has sometimes been found to be of value. The United States Food and Drug Administration has not yet approved the use of cimetidine for treating acid reflux but preliminary results indicate that it may well be of value for this purpose. The mainstays of treatment remain the older standard ones. Small frequent feedings and antacids one or two hours after meals and at bedtime should be prescribed. The head of the bed should be elevated three or four inches. Bending over from the waist, tight belts and garments around the abdomen, and eating just before lying down should all be avoided. Hiatus hernia only rarely requires surgical intervention. If esophageal stricture develops, dilatation with bougies or Eder-Puestow dilators is often successful.

The incidence of carcinoma of the esophagus increases with increasing age and occurs twice as often in men as in women. Treatment with radiotherapy sometimes produces results as good as with surgery and in elderly patients, with major medical contraindications to surgery, is usually the treatment of choice. A Celestin tube, positioned under fiberoptic guidance, can sometimes be used in patients with inoperable malignancy (12).

Stomach and Duodenum

The overall incidence of peptic ulcer disease does not increase with aging, but the distribution between gastric and duodenal ulcers changes as does the sex ratio. The ratio of duodenal to gastric ulcer is two to one in the aged compared to ten to one in the young. The ratio of males to females in duodenal ulcer is five to one in older patients rather than eleven to one in younger ones. In geriatric patients with gastric ulcer, the sexual distribution is about equal. Symptoms of ulcer disease in older patients are often milder and less specific than in younger ones. A perforated duodenal ulcer may appear with a much less dramatic picture in the aged. Gastric ulcer is often seen with vague upper abdominal pain, weight loss, anorexia, nausea and vomiting. When the upper gastrointestinal series reveals even an apparently benign gastric ulcer, the possibility of malignancy must be excluded. Fiberoptic endoscopy with biopsy is a rapid and safe method for differentiation, but following the ulcer radiologically to complete healing with therapy is still sometimes employed. The incidence of bleeding from ulcers does not increase with age, but mortality from bleeding does, so that surgery is often indicated, especially with gastric ulcers. Prompt fiberoptic esophagogastroduodenoscopy has become extensively used, even in the elderly, for diagnosing the cause of significant upper gastrointestinal bleeding and thus helps in deciding therapy. Studies are now underway to determine if the prompt endoscopic diagnosis alters outcome.

Medical therapy of peptic ulcer has changed considerably in recent years. The traditional bland ulcer diet has been found to be unnecessary. The patient is now merely instructed to avoid gastric secretagogues such as alcohol, tea and coffee as well as the foods that produce symptoms in that individual. Decaffeinated coffee has been found to be almost as potent a stimulus of gastric acid secretion as regular coffee. Cimetidine has been a major advance in the treatment of ulcer disease, but it is still too soon to fully evaluate its final role. It may make "intractable" ulcers less common, thus reducing the need for surgery or for resorting to gastric irradiation for complicated peptic ulcers in geriatric patients. Mental confusion is one side effect of cimetidine that occurs almost exclusively in the aged, but it develops only rarely and clears with cessation of the drug. Antacids, used in adequate doses at least four times a day, one hour following meals and at bedtime remains basic for ulcer therapy and works well in conjunction with cimetidine.

The recent marked decrease in the sodium content of the most popular antacids is helpful in those geriatric patients with illnesses that require restriction of sodium intake. Anticholinergics are rarely, if ever, indicated in the treatment of ulcers and they are particularly hazardous in the elderly, who have an increased incidence of benign prostatic hypertrophy in men and of glaucoma in both sexes.

The aging gastric mucosa may be more susceptible to acute gastritis produced by agents such as alcohol, the toxins of food poisoning and ulcerogenic drugs, including aspirin. Removal of the offending agent and simple symptomatic therapy usually results in prompt reversal of the process. If the gradual mucosal atrophy, which occurs in most people with aging, proceeds far enough, it results in chronic atrophic gastritis. The latter often has associated intestinal metaplasia and minimal inflammatory cell infiltration. It is usually asymptomatic or has only mild nonspecific symptoms. There is no specific treatment. It is of importance because it is associated with a very much increased incidence of gastric carcinoma, a disease that increases in incidence with increasing age and is one of the most common malignancies in the elderly. There is some evidence that the incidence of carcinoma of the stomach is decreasing in all age groups in the United States, but the reason for this decrease is not clear.

Small and Large Intestines

Bowel function is frequently disturbed in the elderly and the cause of the disturbance is often not organic (14). Although the constipation or diarrhea may well turn out to be functional in origin, the etiology should be investigated if the symptom is persistent, or if the onset is suggestive of a more serious cause. The techniques for study are essentially the same in the aged as for the younger age groups, but some are more difficult to use in the former. Proper positioning for sigmoidoscopy and retention of the barium enemas are two common difficulties.

Constipation is extremely prevalent in the aged and is often a cause of concern to the patient. The findings on history and physical examination including careful palpation of the abdomen and rectal examination as well as the results of simple routine studies, such as fecal occult blood and hemoglobin, often indicate a need for more extensive investigation, especially if there is rectal bleeding. The most frequent cause is simple functional constipation and, in the aged, is often associated with loss of

bowel tone, change in rectal awareness, decreased mobility, and insufficient dietary fiber and bulk. Other functional causes include motility disorders such as slow transit time with hypotonia, the irritable bowel syndrome (spastic colon), dyschezia and psychiatric illnesses. Drug therapy with narcotics in their various forms, some nonnarcotic analgesics, antacids containing aluminum hydroxide, and some antidepressants with anticholinergic effects may all induce constipation or aggravate a preexisting one. Many organic diseases produce constipation including local rectal lesions such as fissure, fistula, or stricture. Carcinoma of the colon and rectum is the most feared organic cause but others, including diverticular disease and sigmoid volvulus, are also of major importance. Neurologic illnesses resulting in paraplegia and endocrine diseases such as hypothyroidism may also produce significant constipation.

Treatment of chronic constipation in the elderly is often frustrating to the patient and the physician. Many patients have an unrealistic picture of normality and this is reinforced by mass media commercials. Some patients also have a fear of the absorption of "poisons" and "toxins" from the bowels if they are not evacuated thoroughly and frequently. When the elderly patient first consults the physician because of constipation, laxative dependency and abuse is almost invariably present. It is rarely curable but a concerted effort by the doctor usually produces marked amelioration. The patient should be instructed in an appropriate diet with increased fiber such as unprocessed bran, raw fruits and vegetables, and prunes (15). Fluid intake must be adequate. Increased physical activity and exercise, if practical and safe, is also advisable. A regular time should be set aside, preferably after a meal, to take advantage of what remains of the gastrocolic reflex, for the patient to sit on the toilet without tension. At the beginning, a hypertonic phosphate enema or glycerine suppository may be used for a limited period of time to help improve the bowel pattern. Despite all of this it is frequently and, indeed, usually necessary to resort to some sort of aperient, often for an indefinite period of time. The classes include a) bulking agents such as psyllium hydrophilic mucilloids; b) stool softeners such as dioctyl sodium sulfosuccinate; c) saline laxatives or hyperosmotics such as magnesium salts and lactulose; and d) irritating or stimulating laxatives including senna and cascara and polyphenolic laxatives including bisacodyl and phenolphthalein. They should be used in the lowest possible doses for the shortest possible periods of time. Saline

laxatives should be avoided in patients with impaired renal function. Senna and cascara when used in high doses for prolonged periods of time may damage the ganglion cells of Auerbach's plexus and produce colonic dilatation, mimicking Hirschsprung's disease.

Severe untreated constipation may go on to fecal impaction, which may present with paradoxical diarrhea and fecal incontinence or the passage of small putty-like stools as well as tenesmus, obstipation, or even bowel obstruction. It is a frequent problem in some geriatric institutions and is largely preventable if sufficient attention is paid by the staff to the patient's bowel activity (16). Manual disimpaction, unpleasant as it may be, is still the initial mode of therapy, although purgatives, suppositories and enemas may be used after the major impaction has been removed.

Diarrhea is a common symptom in the aged although less common than constipation. It has been frequently emphasized (14) that the approach to diarrhea should depend on the acuteness of its onset, the response to early symptomatic treatment and an assessment of the most likely etiologies. Febrile diarrhea of acute onset is usually infectious in origin. The gradual onset of diarrhea with bulky foul smelling stools is suggestive of malabsorption or maldigestion of small bowel or pancreatic origin. Elderly patients with diarrhea, especially if severe and accompanied by fever and anorexia, tend to become dehydrated quickly and may require hospitalization for rehydration and correction of electrolyte imbalances. It is usually appropriate to treat mild diarrheas of recent onset symptomatically for a few days. If the diarrhea persists, further evaluation of etiology should be undertaken. Complete blood count, stool examination for ova, parasites, culture, and occult blood, sigmoidoscopy, barium enema, upper gastrointestinal series with small bowel follow through and standard blood chemistry profile is often indicated and usually establishes the diagnosis. If not, they may suggest the need for other more specialized tests, such as small bowel biopsy, quantitative fecal fat or pancreatic secretion, to establish or confirm the diagnosis.

Treatment of diarrhea depends on the etiology and severity. When available, specific therapy for the underlying disease process should obviously be used. However, many of these patients as well as the larger number of patients with functional diarrhea or those with conditions for which no specific treatment is available will require symptomatic treatment. Foods that are correlated by the patient with exacerbation should be eliminated as should

naturally laxating foods such as raw fruits and vegetables and prunes. Kaolin and pectin combinations are only minimally effective except in mild cases, but may often be prescribed, almost as a placebo, when it is felt that the acute diarrhea will be self-limited and it is safer to withhold more potent agents. Aluminum hydroxide (without added magnesium salts) is effective in mild cases. In more severe cases, constipating agents such as diphenoxylate and atropine, codeine, and loperamide are quite effective. The older liquid preparations such as deodorized tincture of opium and camphorated tincture of opium are also quite effective and in many geriatric patients have the advantage of permitting greater flexibility of dosage. Antispasmodics may also be required to help control associated abdominal cramps, but, as previously noted, great care must be taken in their use.

Fecal incontinence is also a problem in many geriatric institutions. It is usually caused by underlying bowel disease, fecal impaction, or neurogenic changes. The bowel disease may range from carcinoma of the rectum and severe diverticular disease to anal sphincter damage and rectal prolapse. In this group, treatment should also be directed toward the primary disease. Neurogenic fecal incontinence is thought to be caused by altered neural responsiveness, so that rectal distention is followed by intrinsic rectal contractions and the inhibition of contraction in the anal sphincter. It is often associated with overt neurologic disorders or organic mental syndromes. Incontinence associated with constipation is usually overflow in nature resulting in the continued leakage of liquid or semi-formed stool. When neurogenic in nature, the incontinence from constipation is treated by emptying the colon mechanically and with laxatives, and then an ongoing regimen to prevent its recurrence. Neurogenic incontinence is best treated by induced constipation and periodic planned evacuation. This can be done by giving constipating agents as needed and inducing a bowel movement once or twice a week with enemas and suppositories.

The normal decreases in gastrointestinal function that occur with aging do not contribute significantly to malabsorption syndromes. If symptoms of malabsorption occur in elderly patients, they should not be dismissed because of old age (9). The presence of malabsorption should first be confirmed by appropriate screening tests. The etiologic diagnosis should then be established in an orderly manner, with emphasis on diseases most likely to be found in the elderly. Effective therapy will then often be available.

Ischemic disease of the bowel occurs less frequently than in many other organs in aged patients with their increased incidence of vascular disease because of the rich anastomoses in the mesenteric circulation and the marginal artery of the colon. Acute obstruction of the superior mesenteric artery or vein produces ischemia of the small bowel and appears as an acute abdominal emergency requiring prompt surgery. Gradual progressive atherosclerotic narrowing of the superior mesenteric artery produces abdominal angina. The pain is usually upper abdominal and postprandial and may be confused with peptic ulcer disease or cholelithiasis. Although uncommon, the presence of an epigastric bruit is helpful. The diagnosis can be definitively established by arteriography but increased risks limit its use for this purpose. Treatment consists of small frequent feedings. Nitroglycerine rarely helps. Ischemic colitis has been recognized more frequently in recent years. There is usually significant disease of the inferior mesenteric artery and superimposed factors such as congestive heart failure or hypotension resulting in low flow states. Abdominal pain and bloody diarrhea are usually the presenting symptoms. Edema and submucosal bleeding result in the typical radiologic finding of thumbprinting. If extensive, gangrene may develop, and emergency surgery is required. Milder cases can be treated with antibiotics and intravenous fluids. Unless stricture ensues, surgery can be avoided. Sparing of the rectum helps differentiate ischemic colitis from inflammatory bowel disease.

Diverticula of the colon are uncommon in the young, rarely being present below the age of 40. They occur in 20 to 30 percent of people over age 60, and in more than 40 percent of those over age 80. They are usually asymptomatic, only occasionally causing rectal bleeding. The diagnosis of their presence is established by barium enema or occasionally by colonoscopy. Diverticula are thought to develop in segments of colons with increased intraluminal pressures. There is epidemiologic, experimental and clinical evidence that high fiber diets increase colonic diameter and decrease the intraluminal pressure resulting in a lowering of the incidence of diverticulosis and diverticulitis. Until recently, a low residue diet was usually prescribed for diverticulosis and many geriatric patients have been on such a diet for years. It may be difficult to convince such patients to now switch to the previously forbidden foods of a high fiber diet. A gradual change to such a diet often meets with more success as does adding bulk producing agents and unprocessed bran. Diverticulitis only develops in a small

number of patients with diverticulosis. The most common symptoms are abdominal pain, constipation and/or diarrhea, nausea and vomiting. Localized abdominal tenderness in the left lower quadrant is the most common physical finding but fever and a palpable mass may be present. The diagnosis is established by barium enema, preferably performed at a quiescent stage. Medical management of diverticulitis usually is successful in all but the most severe cases, when surgery is required. Limitation of oral intake, antibiotics, intravenous fluids and analgesics are the mainstays in the treatment of acute attacks. Cautious use of antispasmodics may relieve some of the pain caused by spasm. Meperidine is preferable to morphine or codeine for pain relief because it produces less increase in spasm or constipation than the latter two agents.

Carcinoma of the colon is predominantly a disease of the aged and seems to be increasing in incidence. Recent extensive experience with colonoscopic polypectomies appears to have demonstrated that carcinoma can develop in benign adenomatous polyps. The distribution of carcinoma along the colon is changing with a higher percentage developing beyond the reach of the examining finger or rigid sigmoidoscope than was formerly reported. A high index of suspicion and more extensive screening of stools for occult blood may lead to discovery early enough to permit surgical care. Fulgeration of rectal carcinoma may provide symptomatic relief for patients with metastases and extensive local disease.

Volvulus of the colon occurs mainly in elderly patients, with the majority occurring over the age of 70. The symptoms of abdominal distention, pain, and constipation may be deceptively mild. Plain x-ray films of the abdomen usually suggest the diagnosis. The volvulus may often be reduced by sigmoidoscopy, fiberoptic colonoscopy or barium enema. Surgery may then become unnecessary unless the volvulus is recurrent.

Biliary Tract, Liver, and Pancreas

The recent technical advances, previously described, have made diagnosing disease of these systems simpler and more precise, especially in the presence of jaundice. Extrahepatic cholestasis can now usually be readily detected, permitting more rapid appropriate treatment. The recently described immunologic tests for viral hepatitis are also becoming more widely available.

The principal diseases of the exocrine pancreas

are carcinoma and pancreatitis. Carcinoma of the pancreas is another of the predominantly geriatric malignancies with apparently increasing incidence (17). Unfortunately, symptoms do not develop until a late stage, so that despite our diagnostic advances, the diagnosis is not established until cure is impossible. Painless obstructive jaundice is the most common feature. Psychiatric symptoms including depression and anxiety are frequently present. Acute pancreatitis is usually associated with alcoholism or cholelithiasis and is accompanied with severe upper abdominal pain, nausea, and vomiting. Serum amylase is still the most useful test in diagnosis. Meticulous management of fluid and electrolyte balance is essential in treatment of the acute attack. Repeated episodes of acute pancreatitis may produce chronic pancreatitis with pain, anorexia, nausea, vomiting, and weight loss. Malabsorption, steatorrhea, and diarrhea are less common in the old than in the young. Serum amylase is usually normal. Flat film of the abdomen may demonstrate pancreatic calcification. The secretin test, if available, is usually diagnostic. Pancreatic enzyme replacement improves maldigestion and steatorrhea. Treatment of the persistent pain with analgesics and anticholinergics is often unsatisfactory.

Cholelithiasis and cholecystitis are the biliary tract diseases most often seen in the aged. Gallstones occur more often in women than in men. They have been reported in over 25 percent of women aged 60 and over and can be asymptomatic. The consensus of the nonsurgical literature is that elective cholecystectomy for silent gallstones, except in diabetes, should usually be reserved for patients under the age of sixty. Unless there are pressing medical contraindications, the treatment of symptomatic cholelithiasis is surgical. Chenodeoxycholic acid may soon be released for the treatment of gall stones by dissolution of radiolucent gall stones in patients with a functioning gallbladder. The elderly patient who meets these criteria may then be able to avoid surgery. Choledocholithiasis is more common in the geriatric population and may appear with obstructive jaundice but without the typical biliary colic. Treatment is surgical. The increasing availability of fiberoptic endoscopic papillotomy or sphincterotomy to remove retained stones at the lower end of the common bile duct is particularly valuable in geriatric patients who are poor risks for surgery. Recurrent or persistent acute cholecystitis is often an indication for surgery, but the timing of the latter remains a subject of debate.

Jaundice in the aged is produced by malignant or benign biliary obstruction in about 80 percent of

cases. Since nonhepatic diseases such as hemolytic anemia can also produce jaundice, hepatocellular disease is the cause of jaundice in only about 16 percent of cases in elderly patients. Infectious hepatitis is less frequent in the geriatric age range but tends to be more severe and has a higher mortality rate. Older patients in general take more drugs than younger ones, and they are more likely to develop hepatotoxicity. A meticulous drug history is crucial for evaluating the etiology of hepatic disorders. The usual forms of cirrhosis occur in the aged and their management is essentially the same as in the young. Primary carcinoma of the liver is rare. However, the liver is a very common site for metastases from many primary malignancies. Sonography and nuclear scanning usually are highly suggestive of the correct diagnosis, which can then be confirmed by liver biopsy.

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Hematologic Problems of the Aged

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Within the past ten years, a number of excellent reviews (1-3) have been published describing various hematologic abnormalities in the aged. Some of these manuscripts although quite detailed do not critically weigh the results of isolated observations. In the following article, emphasis will be on the practical application of the material discussed. For the purpose of organization, the relationship of aging to changes in the erythroid, myeloid, and coagulation areas will be presented in separate sections.

Erythroid Elements

The level of hemoglobin in the aged that separates a normal from an abnormal measurement has been the subject of over 50 reports. Some claim that the hemoglobin levels in elderly and young adults are similar, while others find a one to two gram percent difference. The problem of sampling that relates to chronologic age may not parallel physiologic age; so that a well-nourished, active businessman of 75 years of age cannot be compared to a sedentary, infirmed geriatric home resident of the same age. In support of this, Hawkins et al (4) have reported a 1.2 gm% higher hemoglobin in an age-comparable active group as opposed to a sedentary group.

However, subjects of similar background of any age may not show any difference in the amount of hemoglobin, as pointed out by Elwood et al (5), who concluded that the mean hemoglobin level in a large number of English males and females over the age of 65 was within the normal range. This interpretation is based on the World Health Organization (WHO) normal values for men with hemoglobin of 14 gm% and PCV 42% as compared to a hemoglobin of 12 gm% and PCV 35% for women (6). Williams and Nixon (7) analysed their data on patients over the age of 75 seen for routine general practice

examinations and came to the same conclusion. Pincherle and Shanks (8) followed over 2,000 businessmen and concluded that the average hemoglobin in these presumably well-nourished individuals over the age of 65 was only 0.32 gm% lower than the hemoglobin obtained in men under 65 years of age.

These reports represent a minority opinion and a majority of the studies demonstrate a difference in the elderly. The difference may be great as seen in the survey taken by Kilpatrick (9) of a normal population in a rural area of Yorkshire, England. He noted that if a hemoglobin of 12.5 gm% was defined as the lowest limit of normal in males and 12.0 gm% as the lowest limit of normal in females, then 20% of the males and 21% of the females over the age of 65 had abnormal values. These results were restated by Myers et al (10) who determined, in a population of 65 years of age, 55% of the males had a hemoglobin less than 14 gm% and 12% of the females had a hemoglobin less than 12 gm%. If 12 gm% was viewed as normal hemoglobin for males then only 14% of subjects had abnormally low values.

A normal physiologic level in geriatric individuals may be lower than that determined as a normal for younger patients. In studies that measure elderly subjects in two groups, patients over 75 almost without exception exhibit a greater number of individuals with a lower hemoglobin level as compared to subjects in the 65 to 74-year-old group. This difference is not because of an increased incidence of disease. Secondly, normal patients studied in geriatric homes have lower levels of hemoglobin, as compared to those individuals who are of the same age but able to function in the community. The implication is that the geriatric home residents are more physiologically advanced as reflected by their requirements for custodial care. In our experience at the Jewish Home and Hospital for the Aged, a hemoglobin below the WHO normal value is often seen. In the absence of any discernible hematologic disease or other disorder, one then has

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a choice of either describing the patient as normal for age or as a patient with a simple chronic anemia. The latter implies a pathologic condition that may not be valid. In men, a hemoglobin below 11.5 gm% is regarded as abnormal while a value between 11.5 gm% and 12.5 gm% is considered in the indeterminant range. In women, a value below 11.0 gm% is regarded as abnormal and a measurement between 11.0 gm% and 12.0 gm% is considered indeterminant.

In an attempt to explain the anemia of senescence, Takaku (11) studied red cell production and circulation in normal aged people with radioactive labeling techniques. He found slightly elevated values for plasma iron turnover of 28.8 ± 4.4 mg/24 hours and slightly decreased values of 21.0 ± 1.6 days for chromium-labeled red cell survival. These data suggest anemia in the elderly on the basis of inefficient production and decreased survival. Bone marrow scans based on iron isotopic labeling of erythroid elements indicate that active marrow diminishes as age advances. This involution begins first in the long bones and then extends to the smaller flat bones and finally to the vertebrae. As an extreme example, the cellularity of a person over the age of 80 years has been estimated to be approximately 30% of the maximum marrow cellularity present during the first decade (12). The decrease is proportionately the same for both erythroid and myeloid elements so that the cellular composition is unchanged. From a practical view point, it is fairly common for a sternal bone marrow aspiration to be hypocellular. This reflects involution of the marrow rather than overall hypocellularity and is the reason that the posterior iliac crest site is preferred in elderly patients.

There is some conflicting data from mice experiments about the gradual depletion of the stem cell pool during senescence, but it is generally agreed that there is a decreased ability of stem cells to recover following irradiation or to sustain production in patients with continuous blood loss. In Harrison's study (13), red cell production in old mice in response to bleeding was normal through the fourth day, but decreased significantly after this point. Young mice recovered rapidly over a nine to ten-day period. Compatible marrow taken from young mice and given to old mice failed to improve the erythroid production which suggests that a defect in the aged may not be a fault of the stem cell but perhaps an abnormality in the hemopoietic micro-environment. Hematopoiesis is regulated in part by a humoral substance erythropoietin but it is difficult to make any statement about levels of this substance in the elderly since reproducible mea-

surements at the low levels seen at normal hemoglobin values are difficult to obtain. The fall of hemoglobin in geriatric men is believed by some to be the result of hyposecretion of androgens but this is unproved.

Under normal conditions, the aging red cell undergoes changes that originate with decreased levels of various enzymes of intermediate metabolism, notably aldolase, hexokinase glucose-6-phosphate dehydrogenase, and sulfhydryl enzymes like glutathione. As a result, less 2-3 diphosphoglyceraldehyde (2-3 DPG) and adenosine triphosphate (ATP) is generated. Consequently, less energy is supplied by ATP so that the red cell loses its flexible biconcave shape leading to impaired passage through the spleen and cell destruction. Such changes are expected, but abnormally decreased erythrocyte ATP levels have been reported with increasing age in man (14). This may explain the decreased deformability of red cells in a population of people age 69 to 91 years as compared to a control group age 12 to 29 years (15). Similarly, 2-3 DPG levels related to oxygen hemoglobin affinity was reported by Brain and Caird (16) to decrease with age. Some of this data is disputed as shown by Tweeddale et al (17) who could find no significant age variation in the quantity of 2-3 DPG that usually parallels the amount of ATP, in a study of 47 healthy volunteers ranging in age from ten to 90 years. Perhaps the enzyme differences described in the aged are related to the shortened red cell survival.

A comprehensive review of the different types of anemia will not be attempted in the following section but comments will be directed toward those situations that have particular relevance to the elderly. The signs and symptoms of anemia are the same as would be expected at any age but the emphasis is different. Exertional dyspnea, which may be seen clearly in a young adult without any associated disease, may be difficult to evaluate in an elderly patient with pre-existing chronic lung or cardiac disease. Pulmonary edema or ankle edema, unusual in younger patients, may be the first sign of anemia in the aged. In older patients angina pectoris, dizziness, buzzing in the ears, mental confusion, or even unexplained apathy can be the initial symptoms of anemia. Skin color is a poor way to judge anemia in the elderly and attention should be focused on nailbeds and buccal mucosa. Hemicardiac murmurs usually represent turbulence over rigid valves and are common even at normal hemoglobin levels.

Different types of anemia are more prominent at different ages as shown in the following Table (Ref. 3, modified p. 158).

TABLE I

		Age 20-29	Age 40-49	Age over 60
Iron deficiency		20.6%	10.1%	12.3%
Megaloblastic anemia		—	0.7%	3.2%
Aplastic anemia		3.3%	4.0%	0.7%
Hemolytic anemia		1.3%	—	—
Leukemia lymphoma		13.9%	5.3%	2.7%
	Infection-inflammation	29.5%	18.2%	23.4%
	Renal disease	11.9%	8.1%	4.5%
Anemia of chronic disease	Liver disease	3.3%	6.0%	3.2%
or	Endocrine diseases	2.7%	3.4%	2.0%
Simple chronic anemia	Solid tumor malignancy	6.6%	20.8%	35.1%
	Miscellaneous (refractory and sideroblastic anemia)	11.9%	15.4%	13.0%

This Table represents the proportion of different types of anemia in each age group and does not mean, for example, that iron deficiency is less common in the elderly. It is important to note that over two thirds of the patients do not have a primary anemia but have an anemia of chronic disease. A nutritional deficiency of iron, vitamin B12 or folic acid is responsible for fewer than 16% of the anemias in patients over the age of 60 years.

One of the most common differential diagnostic problems in the elderly is to distinguish between iron deficiency anemia and an anemia associated with chronic disease sometimes referred to as refractory anemia. The textbook description of normochromic, normocytic red cells in chronic refractory anemia and hypochromic microcytic cells in iron deficiency anemia is not always followed since both types of anemia may exhibit hypochromia and, to a lesser extent, microcytosis. Iron deficiency is characterized by a low serum iron and an elevated total iron binding capacity (TIBC). An elevated TIBC is seen only in iron deficiency and in situations in which a general total increase in globulin is found as in pregnancy. A patient with a serum iron to TIBC ratio of 16% or less is considered to be iron deficient. Conversely, in anemia of chronic disease a low serum iron accompanied by a normal or low TIBC reflects defective utilization of iron secondary to a block at the reticuloendothelial level. In a rarer type of defective utilization the serum iron is elevated but the TIBC is normal or low. In the latter, sideroblastic anemia iron is frozen in the normoblasts and is not incorporated into the mature red cells.

The problem is that iron levels are lower in older subjects since they tend to fall progressively after the age of 30 (18). Although the mean change is small, the range of normal values is much wider than in younger individuals. Similarly, the TIBC reaches a maximum at age 50 to 60 and declines

thereafter (19). Here again, as shown by MacLennan et al (20) in their study of 188 subjects in a home for the elderly, the problem lay in the wide normal range of the TIBC which varied from 214 mg% to 480 mg%. Thus, a calculated serum iron to TIBC ratio must be judged carefully since it may be subjected to a double error.

If the etiology of the anemia is not clinically obvious, further laboratory measurements can be obtained. The high degree of osteoporosis seen in the elderly make a bone marrow aspirate a quick and easily tolerated procedure. The absence of stainable iron in a deficiency state and the presence of iron in anemia of chronic disease can be accurately assessed. Iron determinations should be made from bone marrow aspirate slides and not from bone marrow biopsy sections, since iron may be removed during histologic preparation of the latter. As an alternate to a bone marrow aspirate, one can measure serum ferritin since one microgram of ferritin per 1000 cc of serum is equivalent to 8 mg of storage iron. However, the determination of ferritin involves measurement of microscopic amounts of material and as such is subject to greater error than histochemical staining of the bone marrow. If any doubt still remains about the presence of iron stores, the patient can be given a radioactive tracer dose of iron. A normal $T_{1/2}$ clearance of 60 to 90 minutes is greatly shortened in iron deficiency.

Simple Chronic Anemia

As described earlier, an anemia secondary to an underlying disease is most common in the elderly. Treatment should consist of transfusion with packed cells if clinically indicated. Since there is no evidence of a nutritional deficiency, hematinics will not raise the hemoglobin level. Androgens can enhance erythroid production and have been used to

raise marginally the hemoglobin level and to decrease the transfusion requirement. Unfortunately, these hormonal preparations are poorly tolerated in the geriatric population because of salt retention leading to fluid accumulation and compromised cardiac status. These drugs also may exacerbate prostatic urinary retention and may produce unpleasant virilism in female subjects.

Iron Deficiency

Iron deficiency is second only to anemia of chronic disease in the elderly and most often is the result of vaginal or gastrointestinal (GI) blood loss. A hiatus hernia is common in this group and is the most frequent source of obvious or occult GI bleeding. Aged patients are more likely to use acetylsalicylic acid or other anti-inflammatory drugs for treatment of arthritis and should be observed carefully for signs of gastritis and ensuing GI hemorrhage. GI arteriovenous fistulas or telangiectasias of the Osler-Weber-Rendu type are more common in the elderly and increase in frequency and severity of bleeding with advancing age (21).

Iron is absorbed in a gradient that is maximal in the duodenum and minimal in the ileum. In patients with sprue or with rapid passage of food following a total or hemigastrectomy, iron absorption is impaired. In the latter, it is probable that not only the rapid passage of food but also the loss of gastroferitin in the stomach contributes to the iron malabsorption. Iron deficiency caused by malabsorption of gastric and intestinal mucosal atrophy can be reversed by iron replacement. Assuming no malabsorption or hemorrhage, it is unlikely that a geriatric subject on a "tea and toast" diet will become iron deficient for the following reasons. Iron is conserved with great efficiency in the body and the amount released from destroyed red cells is recycled for use again. Loss of iron in urine, sweat and hair is insignificant. Each day, approximately two cc of blood containing one mg of elemental iron is lost in the stool which constitutes the daily mineral requirement. Thus, it would take approximately three years to deplete the normal 1000 mg iron stores assuming no iron intake. Even a drastic reduction in the average North American diet of 10 to 15 mg of elemental iron would be balanced by enhanced intestinal absorption. Under normal circumstances, only 10% of the available iron is absorbed but, if necessary, this can be increased up to 40%. This means that even if the dietary intake is reduced to 2.5 mg of iron enhanced absorption ($2.5 \text{ mg} \times 40\%$) will achieve the requirements of one mg

of iron per day. These calculations presume that iron absorption is not impeded by phytates and phosphates in the diet. Nevertheless, in an adult male and in a postmenopausal female one should regard iron deficiency on a nutritional basis as unlikely and proceed on the assumption that there is another reason for the iron depletion.

The administration of iron given as tablets in the ferrous form is preferred since they are absorbed three to seven times more readily than those in ferric form. The medication is most effective when given one hour before meals since food can bind iron and retard its absorption. Oral treatment sometimes fails because of malabsorption but more commonly because of GI intolerance leading to failure to take the prescribed tablets. In this situation, it may be necessary to give parenteral iron. The reticulocyte count begins only one day earlier with parenteral as opposed to oral iron so that a speedier response should not be a criteria for the use of the intramuscular or intravenous preparations. Both the iron-dextran (Imferon) preparation and the iron-sorbitol (Jectofer) preparation contain 50 mg of iron per ml but the former is preferred because of the fewer side effects and absence of a metallic taste in the mouth (22). The quantity of parenteral iron needed is easily calculated assuming that 1 cc of packed cells contains 1 mg of elemental iron and 1 cc of Imferon contains 50 mg of iron. The amount needed should be that required to raise the hemoglobin to normal plus an additional 1000 mg to replenish the tissue stores.

Imferon given by the intramuscular route usually requires repeated injections that may prove to be a problem in an elderly patient who is unable to reappear for successive treatment. To obviate this, Wright (23) and others have advocated administering the total dose of iron as Imferon or as Dextriferron in one liter of saline. The solution is started at 10 drops and then advanced to 40 drops per minute if tolerated. Lasix 40 mg is usually given simultaneously as a precaution against fluid overload. Patients receiving total dose infusion may experience reactions varying from 0.5% to 6% which may include local irritation, venous spasm, thrombophlebitis, headache, nausea, vomiting, pyrexia, and tachycardia (24). Very rarely hypersensitivity reactions may occur with respiratory distress and cardiovascular collapse. Reactions occur less commonly in very severe iron deficiency presumably due to increased plasma iron binding capacity with prompt protein binding. Significantly, no reactions occurred in the 68 elderly patients treated by Wright (23) and the responses overall were good.

Megaloblastic Anemias

Megaloblastic anemias are characterized by morphologic abnormalities consisting of macrocytic red cells and decreased number of white blood cells and platelets in the peripheral blood. The bone marrow is very cellular and contains a marked increase in erythroid elements a majority of which are abnormal megaloblasts. The decreased nuclear maturation as compared to cytoplasmic growth may be reflected in impaired development of all cells within the body producing an abnormality on tissue biopsy, and the associated pancytopenia as described above. Often, this defect is produced by a lack of vitamin B12 or folic acid but rarely can a similar picture be seen in conditions in which nucleic acid synthesis is defective either through an inherited abnormality (25) (orotic aciduria or foriminotransferase deficiency) or following the chronic administration of some types of antineoplastic chemotherapy. Occasionally a megaloblastic bone marrow examination may reflect an early myelocytic or erythrocytic leukemia (Digugliemo syndrome) (26). The incidence of megaloblastic anemia in the elderly is less than 5% of the total number of patients with anemia rather than a figure of almost 50% claimed by Evans et al (27). Nevertheless, decreased levels of vitamin B12 and folic acid represent an important nutritional deficiency state in the elderly.

Vitamin B12 Deficiency

Animal protein in a diet supplies the usual daily requirements of 1 to 2 ug of vitamin B12 which balances the daily loss of 0.03% of the body stores. Intrinsic factor (IF) supplied by the gastric parietal cells combines with vitamin B12 for maximal absorption in the ileum. As much as 5 ug of vitamin B12 can pass through the enterohepatic circulation daily so that lack of IF will produce vitamin B12 deficiency much more rapidly than a dietary deficiency. Assuming body stores of vitamin B12 of 5000 ug, it would take over ten years to produce a deficiency on the basis of impaired nutrition. Unless the patient is a food faddist, even the most meager diet will supply some animal protein containing B12 so that irrespective of age, malabsorption rather than diet is the etiology of vitamin B12 deficiency.

A progressive fall in serum B12 measurements with advancing age has been described by many authors (28, 29) implying that the reduction is caused by impaired vitamin B12 absorption. Normal serum vitamin B12 values are 200 to 900 pg per ml with measurements from 111 to 199 pg per ml regarded as indeterminant, and values less than 110

pg per ml indicative of a deficiency. A British survey compiled in 1972 showed that 10% of elderly subjects had serum B12 levels in the indeterminant range (30). Whether these subjects have latent pernicious anemia is not clear since MacLennan (19) reported a small group of patients with a mean vitamin B12 level of 140 pg per ml who subsequently developed obvious disease. The question as to whether or not decreased Vitamin B12 levels in the elderly reflect decreased amounts of IF caused by the occurrence of atrophic gastritis was studied by Glass et al (31) and Chernish et al (32). They reported that B12 absorption in elderly normal individuals was enhanced by giving IF with the oral test dose of vitamin B12. By contrast, Hyams (33) found that elderly and younger subjects could produce an equivalent normal amount of IF under maximum stimulation with carbamyl choline. Ileal absorption of the vitamin B12-IF complex was equally efficient in both groups. Halsted et al (34) have shown that the liver contents of B12 is unaffected by age. Since the liver contains over 90% of the B12, it appears that the elderly do not have depleted stores of this vitamin.

The relationship between subnormal B12 levels and psychiatric abnormality bears some discussion. Droller and Dossett (35) claimed a correlation between subnormal B12 levels and dementia in a significant percentage of patients over the age of 70 admitted to a hospital. Whitehead and Cholan (36) even described a relationship between vitamin B12 deficiency and paranoid delusions. Strachan and Henderson (37) discussed the possibility that screening for B12 deficiency via serum B12 measurement might be worthwhile in psychiatric evaluation of the elderly with normal blood parameters. This is not of practical value as shown by Murphy et al (38) who obtained serum vitamin B12 measurements on 1004 consecutive new psychiatric patients over the age of 50 and found only two cases of pernicious anemia, an incidence lower than expected. In fulminant megaloblastosis, mental changes may occur associated with severe anemia and, as such, a simple blood count should provide the appropriate laboratory screening test. In the absence of anemia there is no association between mental symptoms and low serum B12 levels.

Folic Acid Deficiency

The usual western diet contains enough fresh green vegetables, fresh fruit, nuts or liver to provide 500 to 1000 ug of folate per day. Under normal circumstances approximately 200 ug of folate is

absorbed generously exceeding the average daily 50 to 100 ug. vitamin requirement. Serum folate levels are difficult to evaluate and low determinations without anemia are common in the elderly. Folate levels below 3 ng per ml are considered abnormal according to standards set by the World Health Organization (39). This figure is considered to be too high by some because of surveys that have identified a putative deficiency in an excessive number of normal aged individuals. For example, the British Department of Health & Social Security (40) determined that 14.6% of 653 elderly subjects had serum folate levels below 3 ng per ml, while Sheridan et al (41) recorded that 30 out of 365 or 8% of normal older individuals had folate levels below 2 ng per ml. Hospitalized patients may have even lower measurements as shown by Buxton et al (42) who found folate levels below 1.5 ng per ml in 27 out of 40 mentally abnormal geriatric patients and 19 out of 40 mentally normal older control patients. These figures were of interest to Sneath et al (43) who found a mean lower red cell folate in the 14 patients in their series with dementia. However MacLennan (20) could find no relationship between folate deficiency and dementia.

The poor correlation between folate levels above 1.5 ng per ml and anemia raises the question as to whether or not the serum levels reflect the body content of folate stored mainly in the liver in the form of 5-methyl tetrahydrofolate. Measurement by liver biopsy can be obtained but measurement of red cell folate (RCF) is more practical since the folate remains in these cells throughout their lifespan (44). Normal values are 150 to 600 ng per ml of packed cells. Measurement of RCF does not appear to be more precise since various studies (40) have reported that 8% to 16% of normal geriatric patients have values below 150 ng per ml.

Despite the likelihood of an inadequate diet, megaloblastic anemia from folate deficiency is not common in the aged (45). Excessive ingestion of alcohol which produces a toxic effect on the bone marrow and is associated with a diet low in folate is not a problem in the elderly. Increased utilization because of increased metabolism with red blood cell hemolysis, during tumor growth or with high activity in inflammatory lesions may severely deplete folic acid but only rarely produces megaloblastic changes in the young or elderly. Clinical states associated with disease of the jejunum producing malabsorption are not more frequent in the older patients. Vitamin C appears to help maintain folic coenzyme in a reduced state and facilitate its absorption although the relationship between these

two vitamins is unclear. Usually, the anemia in a scorbitic patient is the result of tissue bleeding and is characterized by normoblastic erythroid hyperplasia. However a deficiency of Vitamin C in an elderly, poorly nourished patient with borderline folate stores may rarely produce a megaloblastic bone marrow (46). In the absence of malabsorption, 1 mg of folate given by mouth three times per day is recommended as the initial treatment. Subsequently a maintenance dose of 1 mg of folate per day provides a sufficient amount of this vitamin.

Erythroleukemia (Di Guglielmo Syndrome)

A megaloblastic marrow which does not convert to normal erythropoiesis after the administration of vitamin B12 or folic acid suggests an erythroleukemia syndrome with ultimate evolution into an acute myeloblastic leukemia (47). Erythroid precursors show similar disturbances as in B12 or folate deficiency namely a slowing of DNA synthesis and a lengthening of the synthetic(S) phase of the cell cycle. Often a distinction between a nutritional deficiency and a Di Guglielmo syndrome can not be made on the marrow examination but sometimes nuclear budding and duplication of the red blood cells and an increased number of myeloblasts in the white cell series indicates the probable diagnosis of early acute leukemia. As will be described in the white cell section, an indolent progression particularly in the elderly, may mark the transition to a neoplastic blood dyscrasia. During this period, packed red cell transfusions and observation may be the only treatment required.

Aplastic and Hypoplastic Anemia

Aplastic anemia is uncommon in the elderly. In the series of Evans et al (48), it represented less than 0.2% of hospitalized patients over the age of 65. The diagnosis must be confirmed by bone marrow biopsy to avoid the occasional confusion produced by a hypocellular bone marrow aspirate obtained from patients with aplastic anemia and from those with aleukemic leukemia. In the former, the aspirate is legitimately hypocellular but, in the latter, a "packed" marrow produces technical difficulty in obtaining a specimen resulting in an artificially sparse marrow. A delayed effect from prior exposure to industrial pollutants is possible, but an increased use of multiple drugs in the aged creates a more common potential for aplasia of the marrow. Many drugs have been linked with marrow aplasia (49), but gold salts, phenylbutazone, and chloramphenicol have produced the highest number of

blood dyscrasias. Elderly patients withstand complications of infection and bleeding less well and recovery is less likely as compared to children or young adults.

Senile hypoplastic anemia, an uncommon indolent disorder affecting more women than men (50), is characterized by a decrease in erythroid elements in the marrow creating a failure of erythropoiesis. Unlike pure red cell aplasia, there is no marked lymphocytic infiltration and erythroid precursors are present in reduced numbers. The initial anemia may be followed some months later by leukopenia and thrombocytopenia reflecting a decrease in myeloid and megakaryocytic elements. Treatment with androgens may provide slight benefit but packed cell transfusions provide the major treatment.

Serum Globulin and Antibody Levels

There are conflicting reports, but the general consensus is that gamma globulin levels rise in the elderly up to the seventh decade and then fall with advancing age (51, 52). A wide variety of conditions that can cause this small increase in globulin range from chronic infection or inflammatory disorders to the presence of neoplastic disease. In general, in the aged IgG and IgA levels rise while IgM levels remain the same or decline (53, 54). This is independent of a consistent age related increase in the incidence of IgG auto antibodies (55).

An indication that the immune system may undergo senescence derives from information on age related changes in blood group antibodies. As early as 1929, Thomsen and Kettel (56) reported that anti A and anti B iso-agglutinin blood titers rise rapidly to a peak level about ten years of age and then decline gradually with advancing age. In individuals over the age of 65, the titers are 25% of the peak level. Studies on mice that are believed to parallel the experience in man have shown that the decreased activity in the immune system in elderly mice is caused not only by a decrease in the activity of the immunocompetent cell population, but also by environmental changes that are not conducive to maximal expression of these cells. This information is derived by measuring the activity of spleen cells from normal adult syngeneic donors transplanted into irradiated young and aged recipient mice (57).

Myelolymphocytic Elements

Both qualitative and quantitative changes in white cells may occur with aging. Caird and others

(58) have reported that the mean white blood counts of 500 elderly normal people were leukopenic and lymphocytopenic as compared to younger subjects. The lower limit of normal in their study was 3000 cells per cu mm and is consistent with our unpublished observation that the elderly may have a physiologic decrease in the white blood count. These statistics are based on analysis of white patients and are not influenced by the lower counts seen frequently in black patients of all ages. White blood counts obtained in the early morning can vary as much as 1000 cells per cu mm compared to blood counts obtained in the evening. Perhaps this diurnal variation may be responsible, in part, for earlier observations that did not show a change of total and differential counts with age.

There seems to be a division of opinion as to whether granulocytes in the aged display any morphological abnormalities. Olbrich (59) described more marked lobulation with less granulation, and Adam et al (60) noted more four lobed nuclei and a higher than normal Arneth-Cooke count. This measure of the amount of nuclear segmentation is based on the number of lobes in 100 granulocytes. Whether the hypersegmentation is a manifestation of age or reflects early megaloblastosis is difficult to determine particularly when Sheridan, Temperly and Gatenby (41) found a significant correlation between polymorphonuclear lobe counts and borderline vitamin B12 and folic acid levels. There seems to be a shift toward anaerobic metabolism in the elderly as measured by decreased transketolase (61) activity reflecting decreased pentose phosphate metabolism. Additional changes in granulocytes in the aged have included an increase in pyruvate kinase (62) and a decrease in leukocyte alkaline phosphatase (LAPA) (63). The magnitude of the LAPA is too small to create any confusion about the diagnosis of chronic myelocytic leukemia.

Under normal circumstances, mature granulocytes have a half life of four to six and one-half hours and remain in the circulation no more than ten hours. Granulocyte kinetic studies comparing results in older and younger subjects are not available in man but studies of duodenal mucosal cells in mice show a cycle length of 11.5 hours in young mice as compared to an average of 15 hours in old (over 940 day) mice (64). The increased cycle length reflects a longer synthetic (S) and initial resting (G_1) phase rather than any alteration of the second resting (G_2) or mitotic (M) phase. The total blood granulocyte pool is composed of cells in circulation and those along the walls of capillaries and venules comprising the marginal granulocyte pool. Under

conditions of stress or following the injection of epinephrine, cells are mobilized from the marginal pool and enter into the circulation raising the granulocyte count. In the elderly, this movement of cells is less and more delayed (65). In addition, the bone marrow reserve, which is a measure of the ability of the marrow to deliver mature granulocytes into the circulation, is also reduced. As a consequence, an older subject may demonstrate impaired leukocytosis in response to infection or the administration of steroids, endotoxin, or etiocholanolone.

A relative lymphopenia produced by a decrease in the number of circulating T cells was stated to be a characteristic of old age by Augener et al (66) but there is a difference of opinion whether the elderly show a statistical depletion in the total number of lymphocytes (67). Actually changes in B and T cell function may be more important than the absolute number of cells. As an example, the B cell population in the spleen of disease-free long lived old mice is the same as in the spleen of young mice (68). There is a small five-fold difference in the number of immunocompetent cells in the spleen of young and aged mice, but a 50-fold difference in antibody response (69). It is clear also from mouse and human data that the decreased number of T cells in the circulation reflects a decrease in the T cell related immunologic responses which begin to decline early in adult life and continue to fall in the geriatric age. This is based on in vitro decreased proliferative responses to mitogens and to the decreased stimulatory effects of allogenic cells in the mixed lymphocyte culture as well as an in vivo decline in cell mediated immunity. The reduction in humoral immune response in the elderly despite normal or only a modest decrease in the B cell population may be due to a functional or absolute decrease in regulator T cells (70). This is particularly significant since decrease in antibody formation is synchronous with thymus involution. Lymphocytes in the elderly do exhibit an average decrease in incorporation of C^{14} -leucine into protein and an increase in chromosomal abnormalities (71) which principally take the form of aneuploidy (72). According to Lisco et al (73), chromosomal abnormalities between the age of 75 to 92 are five times higher than in lymphocytes of individuals with an average age of 35.

Myeloproliferative Disorders

In the last 50 years, there has been an overall increase in the incidence of leukemia which in the

elderly is seen as an increase of chronic lymphocytic and acute nonlymphocytic leukemia. The management of this latter leukemia bears particular comments because of its variable initial presentation. In the elderly, acute nonlymphocytic leukemia begins more commonly as an indolent blood dyscrasia characterized by a variable picture of anemia associated with either or both leukopenia and thrombocytopenia (74). An elevated reticulocyte count may be seen with occasional macro-ovalocytic red cells and normoblasts. Often a high percentage of monocytes are present and rarely do granulocytes have a pseudo Pelger-Huet morphology. This pre-leukemic picture may be associated with a bone marrow that shows only erythroid hyperplasia or that demonstrates small focal increase in the number of myeloblasts. Although a certain diagnosis cannot be made until the development of a significant number of myeloblasts, the peripheral blood and bone marrow pattern is so definite that a diagnosis of impending leukemia can be made with a high degree or certainty in most instances (75). A low leukocyte alkaline phosphatase, a slight increase in fetal hemoglobin, and a positive acid hemolysis test associated with peripheral blood cytopenias are additional evidence suggesting a leukemic evolution. More recently, it has been shown that these patients have a decreased colony forming capacity in vitro culture and qualitative abnormalities in colony stimulating activity (76). Fifty to sixty percent of patients have chromosomal abnormalities which may precede the development of leukemia by many months. A careful analysis of cases has shown that leukemia occurred in one half the cases in 12 months and three quarters of the cases in 24 months, but in some instances leukemia did not develop for five or ten years (75).

The overall remission rate for patients with acute nonlymphocytic leukemia with the best combination chemotherapy of cytosine arabinoside and either adriamycin or daunomycin varies between 50% and 80% depending on the individual study (77). This declines to 20% to 50% when patients over the age of 50 are analyzed for remission induction (78). The range is large and patients in the ninth decade have attained remission. Moreover, once in remission the duration is comparable to that established in younger patients (79). The reasons for the difference in tumor remission in the elderly probably are multifactorial and relate to the presence of other disease, or a decrease in immune competence, but mostly to prolonged marrow hypoplasia with ensuing risk for infection. The ability to control patients with indolent or "smoldering"

leukemia with red cells and platelet transfusion and the poorer results in the elderly if cytotoxic drugs are used to induce remission have prompted some leading blood specialists to discourage aggressive treatment (80). On the other hand, Rosner (81) does not believe that the treatment can be denied on the basis of age even though remission percentage is lower. Most hematologists would follow a patient conservatively until infection or risk of bleeding forced a more aggressive approach. At this point, treatment should be selected for those elderly patients with a good performance status.

Acute nonlymphocytic leukemia is only one form of myeloproliferative disorder—which includes polycythemia vera, essential thrombocythemia, myelofibrosis–myeloid metaplasia and chronic myelocytic leukemia. The latter occurs most often in middle age and less frequently in older patients. By contrast, almost all of the 110 patients reported by Boruncle and Doan (82) who had myelofibrosis–myeloid metaplasia were over the age of 40 and most commonly were between the age of 60 to 70 years. In general, management of the chronic myeloproliferative disorders in the elderly is well tolerated and consists of reduction of significantly elevated red blood, white blood or platelet counts. A cohort of patients with chronic myelocytic leukemia who develop chromosomal abnormalities in addition to “Philadelphia” translocation are at great risk for developing acute leukemia. Determining if more aggressive treatment during the early phase of disease will be beneficial is under study at the present time.

Lymphoproliferative Disorders

Blood diseases included in this category are chronic lymphocytic leukemia, Hodgkin's disease, and non-Hodgkin's lymphoma, multiple myeloma and Waldenstrom's macroglobulinemia. All are seen more frequently in the elderly and their incidence increases with age (83). Hodgkin's disease exhibits an initial peak in incidence from 20 to 30 years with a subsequent decrease followed by an increase in incidence to the previous level at age 45 to 50 and an increase with age thereafter (84).

Chronic lymphocytic leukemia appears to be diagnosed more frequently than in the past. This may be due to the increased number of blood counts performed on patients. Identification of early disease is associated with a long asymptomatic existence but patients who have splenomegaly, thrombocytopenia or anemia have an increasingly poorer prognosis. For most patients with early or stable

disease, observation or treatment with an oral alkylating agent is all that is required. The death rate from chronic lymphocytic leukemia has been calculated by Gunz and Hough (85) and ranges from less than 1 per 100,000 in patients under the age of 55 to 11.3 per 100,000 for males and 6.6 per 100,000 for females over the age of 75.

Hodgkin's disease in the elderly is associated with a poorer prognosis than in younger patients. The response to chemotherapy or irradiation is not worse—it simply reflects that, in older patients, Hodgkin's disease is present on the average in a more advanced stage and more commonly has an unfavorable mixed cellular or lymphocyte depleted histology (86). Elderly patients usually do not tolerate combined abdominal-pelvic irradiation and full dose chemotherapy even if given sequentially because of a decreased bone marrow reserve. It may be possible to treat selected debilitated patients with combination chemotherapy but, in general, it is preferable to treat an older population with a regimen containing no more than a single drug rather than a combination of cytotoxic drugs.

Non-Hodgkin's lymphomas are classified into nodular and diffuse categories and then subdivided into well differentiated lymphocytic, poorly differentiated lymphocytic, mixed lymphohistiocytic and pure histiocytic subgroups (87). Patients with nodular lymphomas exclusive of the nodular histiocytic type, or patients with diffuse, well differentiated lymphomas do very well on conservative drug therapy. Treatment of this population with a single alkylating agent may produce a delayed response as compared to high dose combination drug therapy but the morbidity is much reduced and the end results are the same (88). A diagnosis of malignant lymphoma without some attempt to subdivide into further categories is not tenable since single drug therapy is based on accurate pathologic classification. The development of lymphomas at extranodal sites is more common in older patients and requires individual assessment. Primary malignant lymphoma of the elderly breast is commonly noted (89) and responds well to local irradiation.

Multiple myeloma is a fairly frequent neoplastic disease of elderly patients with a peak incidence between the ages of 60 to 70 years (90). Often, back or hip pain which would be handled more aggressively in younger patients is discounted as arthritis. Only when anemia or an elevated erythrocyte sedimentation rate is present is an evaluation undertaken to prove an underlying plasma cell infiltration. The cortex of bones in the elderly are generally softer permitting easy entrance of the aspiration of

biopsy needle. This is particularly true of patients with multiple myeloma in whom sternal marrow aspirations must be performed carefully because decreased resistance signifying passage through the cortex into the marrow may be absent. Weakness of bones may produce collapse of vertebrae with resulting pressure on spinal cord. Signs of cord compression are usually obvious but patients with antecedent impaired ambulation or urinary excretion must be watched carefully to detect early signs of paraplegia. A bone marrow examination that demonstrates large collections of plasma cells establishes the diagnosis but often plasma cells are present in only modest numbers. Aspiration from another site or a bone marrow biopsy may obtain additional marrow containing the focal pathologic material. Monoclonal globulin abnormalities are usually present in the serum but may be absent in patients with selective over production of light chains. Electrophoresis of urine rather than a Bence-Jones heat test is recommended for more accurate measurement of these light chains. Polymerized light chains contribute to one form of amyloidosis which is seen in 10% to 25% of chronically ill elderly patients depending on the series quoted (91).

Multiple myeloma is an indolent disease that does not require therapy in the early stage. When indicated, treatment consists of local irradiation for pain, spinal cord compression or, as an adjuvant to orthopedic management, in impending fractures of long bones. Chemotherapy with equally effective alkeran or cytoxan is the major form of treatment. Prednisone, contrary to the initial studies, does appear to add a slight advantage and if given in a pulse dose every four to six weeks does not produce long term side effects. Combination chemotherapy as advocated by Lee et al (92) does not offer a clear advantage over single drug treatment (93) and is difficult to use in many older patients.

Primary macroglobulinemia (94) is a rare condition of the elderly with a peak incidence between 60 to 80 years. An increased number of lymphocytic cells are found in enlarged lymph nodes, hypertrophied liver or spleen and in the bone marrow. An increase in plasma viscosity, if present, may produce retinopathy, peripheral neuropathy and heart failure. An elevated level of IgM globulin is present in the serum in the classical Waldenstrom disease but polymerized IgG and IgA globulin also can produce a hyper-viscosity syndrome. Treatment consists of either or both an oral alkylating agent like chlorambucil and plasmaphoresis (95). In older patients plasmaphoresis must be executed very carefully to

avoid circulatory imbalance. Infusion of saline used to keep the needle patent during the procedure should be kept to a minimum to avoid salt overload.

Hemorrhagic Disorders

Effective hemostasis requires a sufficient quantity of clotting factor, an adequate quantity and normal function of platelets and an intact vascular tree. Alterations in these categories producing an abnormality of coagulation in the elderly will be described.

Clotting Factor Deficiency

In general, the conventional prothrombin time (PT) and the activated partial thromboplastin time (APTT) designed to screen for a clotting factor abnormality do not differ in the elderly. The APTT is more sensitive than the PT and may be erroneously prolonged in patients with an elevated hematocrit or incorrectly shortened or read as normal in the patient with anemia. Although elderly patients do tend to have lower hematocrits, the levels are usually above 36% so that no correction is required. Analysis of individual clotting factors has produced variable results probably in part the result of technical difficulties of measurement. A majority of coagulation specialists do not think that clotting factors are different in the elderly (96, 97), but Hamilton et al (98) found a decrease in level of factors XI and XII in men but a rise in older women. Factors V, VII and X were noted to rise in moderately advanced age but to fall in the extreme elderly. No real difference in the amount of Factor VIII between the elderly and normal adults has been described. Antithrombin III increases in postmenopausal women (98) but decreases in elderly men along with heparin cofactor activity (99).

Congenital factor deficiencies are usually apparent by the time a patient reaches advanced age. A history of excessive bleeding following dental or other surgery or after a serious accident provides more valuable information than coagulation screening tests. If a congenital defect is revealed in the older patient it is usually a subtle von Willebrand's abnormality (100) or a Factor XI deficiency (101). A patient with von Willebrand's Disease has a decreased Factor VIII antigen which may be associated with either or both decreased Factor VIII level and impaired platelet aggregation with ristocetin. Treatment consists of infusion of plasma which will correct both clotting and platelet abnormality. Factor XI has a half life of approximately

60 hours so that a small amount of plasma can produce effective sustained hemostasis. Acquired clotting deficiencies caused by liver disease, malabsorption, or use of coumarin type anticoagulants are treated with Vitamin K or plasma depending upon the presence of an intra or extra hepatic abnormality.

Antibodies may develop in multiplied transfused patients with absent or low levels of clotting factor. However acquired deficiencies especially of Factors V or VIII, unrelated to transfusion can develop spontaneously in the elderly of either sex as a result of antibody like inhibitors (102). Sometimes acquired factor deficiency develops in association with lupus erythematosus (VIII deficiency), nephrotic syndrome (IX deficiency), or amyloidosis (X deficiency); but in the geriatric population there is usually no associated disease. Why inhibitors of clotting factors should arise in elderly subjects or in women post partum (103) is not understood. Usually there is no spontaneous hemorrhage despite an undetectable level of factor. Fortunately these inhibitors have a transient duration.

The amount of fibrinogen, easily measured by colorimetric or heat precipitation methods, increases with age and may double the value for younger subjects. While albumin globulin and the degree of rouleaux determine the erythrocyte sedimentation rate, it is influenced greatly by the fibrinogen level. This is confirmed by the observation that in general the fibrinogen level and the ESR increase in parallel with advancing age. Gilbertsen (104) in his study of 4000 healthy adults found that 75% of 532 women over the age of 65 had an ESR over 20 mm/hour. The median range of 35 mm/hour for those between 65 to 69 years increased to 53 mm/hour for those 70 to 79 years. Hayes and Stinson (105) noted a slow rise of the ESR until age 60 and subsequently a more rapid increase. On the other hand Pinchele and Shanks (106) have reported a leveling of the sedimentation rate after the age of 60. It is obvious that the interpretation of a moderately elevated ESR with no evidence of any disease is difficult because of the overlap of normal and disease. As an extreme example Liljestrand and Olhagen (107) have followed 21 elderly patients with high ESR over 100 mm/hour without any significant globulin alteration for over 20 years and found them to be free of disease.

Fibrinogen deficiency is an acquired abnormality in the elderly and is the result of impaired formation with end stage liver disease or more likely is due to disseminated intravascular coagulation (DIC) or primary fibrinolysis. Fulminant DIC is associated

with decrease of labile clotting Factors V and VIII and a reduced number of circulating platelets. Unfortunately the diagnosis is not always clear in instances of transient or chronic DIC which may produce moderate thrombocytopenia with only a negligible decrease in labile clotting factors. Fibrinogen levels are normally elevated in the geriatric population and the situation can appear when a significant reduction in the amount of fibrinogen may not be appreciated because the recorded level is a "normal" for a younger subject.

Investigators seem to be divided about the general activity of the fibrinolytic system in the elderly. Reports of increased or decreased activity must be evaluated against studies that show no change in the euglobulin lysis time and other parameters of measurement. Hamilton et al (108), in a detailed study, reported an increase in the principal antiplasmins α -1 antitrypsin and α -2 macroglobulin in the blood. He found that plasminogen activator rose and plasminogen fell after the age of 75. At least in elderly males blood fibrinolytic activity is inversely related to obesity. Elevated plasma fibrin monomers seen in many patients with hyperlipidemia may be from enhancement of platelet aggregation with release of platelet factor 4 leading to in vivo thrombin formation and subsequent breakdown of clot.

Anticoagulation with heparin is of value in elderly patients during and following surgery particularly in those with a history of thrombophlebitis. A minidose regimen consisting of 5000 units given subcutaneously two hours before and then every 12 hours after surgery until effective ambulation will reduce the incidence of thrombo-embolism at the cost of a tolerable increase in wound hematomas. Regulation with a partial thromboplastin time is not required in this setting. For certain operations like total hip replacement the minidose regimen is insufficient and full dose heparin should be considered. Here an adequate even amount of anticoagulant can be assured if heparin is given continuously intravenously preferably with an infusion pump to control the rate rather than intermittently. Monitoring the anticoagulant effect of conventional doses of heparin with the APTT to assure an adequate dosage is useful but has little value in preventing hemorrhage since there is no correlation between the APTT and bleeding except when extremely large quantities of this drug are used. At the present time, studies are in progress comparing the anticoagulant effect of low-dose heparin and coumadin (109). A recent report does indicate fewer complications of bleeding but no advantage of low-dose heparin over couma-

din in patients in preventing acute rethrombosis of deep veins (110). There is no evidence to indicate that small doses of heparin is an effective treatment for a debilitated medical patient on prolonged bed rest. The side effect of heparin (111) namely urticaria, fever, and reversible thrombocytopenia pose no greater problem in the aged but osteoporosis from long term use of this drug is a disadvantage in the elderly patient.

Oral anticoagulation with warfarin type drugs given for long-term outpatient prevention and treatment of thromboembolic disease produces more side effects in the elderly. Skin hemorrhage may be caused by reduced elasticity of tissue in the aged. GI bleeding may be the results of hemorrhage from a commonly present hiatus hernia. As stated earlier, antithrombin III levels are on the average lower in the elderly men but it remains to be proved whether this can be correlated positively or negatively with unexpected bleeding. Some of the large number of drugs taken by these patients have a potential for altering platelet function or interacting directly with coumarin derivatives. In the latter group drugs either displace coumarin derivatives from albumin prolonging the prothrombin time or they accelerate the metabolism of mitochondrial enzymes in the liver leading to enhanced metabolism of the coumarins and a shortened prothrombin time. A few substances directly interfere with absorption of Vitamin K. A list of drug interactions is provided in any recent hematology textbook. Most of the drug interaction is produced by nonsteroidal antiinflammatory drugs and barbiturate sedatives which can be continued provided they are taken absolutely regularly. Under these conditions adjustment in coumarin dosage can be made to keep the prothrombin time in the therapeutic range.

Quantitative and Qualitative Platelet Abnormalities

Except for the study of Banerjee and Etherington (112), platelet counts in the aged are similar to counts obtained in younger subjects. Although the number of circulating thrombocytes is normal, the total number of megakaryocytes in the body is reduced. Even in young adults the capacity of the marrow to regenerate platelets is proportionally less than the capacity to produce white cells. This is further decreased in the elderly because of a reduction in the amount of active bone marrow. Clinically, this may be reflected in severe thrombocytopenia following conventional doses of irradiation or chemotherapy. When confronted by a patient with a low platelet count the first consideration, in a

patient of any age, is to determine whether the thrombocytopenia is the result of lack of production or the consequence of peripheral destruction of thrombocytes. It is particularly important to exclude a drug reaction as the etiology of thrombocytopenia in elderly patients who may be receiving multiple therapeutic agents. Almost all drugs have been reported to cause thrombocytopenia but quinine, quinidine, sulfonamides, antibiotics, oral hypoglycemic agents, barbiturates, digitoxin, chlorothiazide and lasix are the most commonly encountered medications (113). Correction of the thrombocytopenia usually occurs within two weeks following discontinuation of the drug. During initial few days drug induced thrombocytopenia and Idiopathic Thrombocytopenic Purpura (ITP) may be confused. A diagnosis is often established on the basis of serial platelet counts after stopping medication. The acute childhood type of ITP is rare in the elderly who have a more indolent chronic disease, respond less well to corticosteroids, and come more frequently to splenectomy (114).

If the platelet count is normal, a bleeding time and Rump-Leeds measurement can be used to indicate abnormal platelet function. There is no data to indicate any alteration of the bleeding time in healthy geriatric patients. By contrast, the Rump-Leeds test is almost always positive irrespective of platelet function because of increased capillary fragility. Tests of both platelet aggregation and adhesion are subject to great error and data based on these measurements may be regarded with skepticism. Nevertheless, the consensus indicates an increased aggregation with ADP and a decrease or unimpaired response to collagen in the elderly (115). Bentzen et al (116) studied platelet adhesion in 63 patients under psychiatric care and found a wide scatter of results with no age related variation. Adams (117) came to the same conclusion and noted some high values for platelet adhesion in normal older subjects and in stroke patients but no consistent trend.

The high incidence of coronary artery and cerebrovascular occlusion in the geriatric population has stimulated interest in drugs which alter platelet function. Presumably interference with the ability of platelets to adhere to foreign or subendothelial surfaces and the ability to block platelet aggregation will prevent thrombus formation. This result can be achieved in two ways. Aspirin blocks the cyclo oxygenase enzyme inhibiting the formation of prostaglandin endoperoxides and thromboxane A_2 from arachidonic acid. Thus the triggering of platelet aggregation by thromboxane A_2 is prevented. Alter-

natively Dipyridamole (Persantin) inhibits phosphodiesterase slowing the breakdown of cyclic AMP which potentiates the effect of prostacyclin a powerful anti-aggregating substance. The practical application of these drugs is under present investigation in several large cooperative studies (118, 119).

Vascular Purpura

Purpura, which occurs mainly on the extensor surfaces of the forearms and hands in an otherwise normal elderly individual, presents the most common referral to a hematologist for a suspected coagulation problem. In fact Banerjee and Etherington (112) in a random survey of 533 geriatric patients found 18.9% of the males and 10.2% of the females had purpuric areas on the extensor surface of their limbs. There was no relationship between the size or extent of the lesions and the underlying clinical state or the age of the patient in the geriatric group. Platelet function studies in the purpuric and non purpuric groups were basically the same.

The loss of subcutaneous fat and changes in connective tissue permit excessive mobility of vessels and the resulting shearing forces allow rupture of small vessels. This senile purpura and similar steroid induced purpura, has a reddish blue color and sharply demarcated borders. Feinstein et al. (120) was able to induce this type of purpura by subcutaneous injection of autologous red cells in both young and aged men. All subjects demonstrated no evidence of an inflammatory reaction and delayed removal of red cells was uninfluenced by steroids, digesting enzymes or ultraviolet light. Senile purpura is not dangerous and is only a cosmetic problem. Its incidence can be reduced by avoiding drugs like aspirin, antihistamines, phenothiazines or persantin which interfere with platelet function. Vitamin C is often recommended for therapy but studies have shown that senile purpura does not correlate with leukocyte ascorbic acid levels (121).

Vitamin C has been reported by many authors to be low in the plasma and leukocytes of elderly subjects (122). This is principally the result of a reduced intake but may be due to any independent effect of aging. Leukocyte ascorbic acid levels below $15 \mu\text{g} \times 10^8$ white blood cells is considered abnormally low. Scurvy, which may be present in the elderly, vitamin C depleted, poorly nourished patient, is associated with skin purpura and bleeding under the periosteum or in the viscera. The hemorrhagic tendency is the result of impaired collagen synthesis, an endothelial defect in capillaries as well as altered platelet function (123).

The relationship of vitamin C and mortality in the elderly has been studied carefully. Wilson et al (124) showed a significant increase in mortality in hospitalized females with an ascorbic acid level below $15 \mu\text{g} \times 10^8$ leukocytes. Burr et al (125) noted that ascorbic acid levels below this level were associated with an increase in overall mortality particularly in men, that was not influenced by smoking which can alter vitamin C measurements. However prophylactic administration of vitamin C in an initial dosage of 150 mg and later 50 mg per day produced normal vitamin C levels but did not reduce mortality below that of the placebo group. This study is supported by that of Gilmore (126) in which vitamin C given for three years did not lessen the mortality of 300 elderly subjects.

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Terminal Pneumonia in the Aged

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The significant role of pneumonic infection in the termination of life in the elderly has been known for more than a century. Sir William Osler, in his description of the disease, emphasized its virulence for the aged. "No other disease kills from one-fourth to one-third of all persons attacked and so fatal is it that to die of pneumonia in this country is said to be the natural end of the elderly" (1). At the beginning of the twentieth century, the mortality rate from pneumonia was estimated at 1500 per 100,000 population at age 70, with a progressive decline in mortality to about 700 per 100,000 population in the period prior to the introduction of antibiotic therapy (2). More accurate statistics obtained at a later period, when the benefits of antibiotic therapy were well established, present a picture more consonant with present day experiences. The tables of mortality from pneumonia and influenza covering the period 1965 through 1974 issued by the Metropolitan Life Insurance Company (Table I) clearly established that pneumonia mortality had declined significantly in all age groups, except the elderly. Individuals age 75 and over have a mortality rate that is more than five times greater for males and seven times greater for females when compared to the age group 65 to 74. Despite the advent of antibiotic therapy, the attrition of the elderly by the pneumonic process is still marked.

The tables listed below are not supported by anatomic confirmation of illness and, hence, do not present a complete picture of the role of various diseases in the mortal process. To evaluate the role of terminal pneumonia in the aged, the author reviewed the postmortem examinations performed at the Jewish Home and Hospital for Aged in New York City during the past 37 years.

A total of 1696 autopsies were performed from 1943 to the end of 1979. The subjects were all full time residents of the home for varying periods, age 65 and over. During their residence, medical ser-

vices were provided. An attending staff made rounds twice weekly and was on duty for additional consultation. Resident physicians were on duty at all times. In addition to routine medical facilities, roentgenologic services, specialty consultations (cardiology, hematology, pulmonary, neurology, surgical) and supervised nursing care were available. The autopsy rate was variable, ranging from a high of 70% to a low of 25%, more recently in the latter range. Autopsies were performed by a trained pathologist in the author's absence (the author estimated that he performed about 70% of the post-mortems reported).

Forty-one percent of the autopsied patients had anatomical evidence of pneumonia either alone or associated with other anatomical findings of disease. Fifty-nine percent of the patients autopsied were free of pneumonic complications. Only fourteen percent of the deceased at autopsy had moderate to extensive pneumonic inflammation without other gross evidence of disease that might have significantly contributed to death.

The diagnosis of terminal pneumonia was made when it appeared on anatomical evidence to be essential in the mortal process. No doubt the frequent presence of other underlying disease may have played an important role, but the pneumonia was anatomically significant.

The role of age as a determinant in terminal pulmonary disease is not easily assessed. In our institution the average age at death of autopsied cases increased eight years during the 37 years of this study. From 1943 to 1959, the average age of autopsied patients was 78.6 years. In the later periods (1960 to 1979) it rose to 86.4 years. The patients that died with terminal pneumonia showed a similar rise in age at death.

As indicated above, 14% of the autopsied died primarily with pneumonia without significant anatomically demonstrable disease. Howell (3) examined 2221 autopsies in a general hospital in patients 65 years of age and over and reported an incidence of 12.6% of deaths due to fatal respiratory disease. His experience appears similar to ours.

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TABLE I
Mortality from Influenza and Pneumonia, 1965-1974

AGE	Death Rate per 100,000										% decrease 1965-1974
	MALE										
	1965	1966	1967	1968	1969	1970	1971	1972	1973	1974	
65-74	126.5	128.5	114.2	146.7	135.8	123.1	103.0	119.5	113.4	95.0	25
75 and over	516.7	518.7	473.9	561.6	522.6	488.3	478.9	527.7	549.9	498.6	4

FEMALE											
65-74	57.0	59.9	51.2	66.9	62.6	55.2	48.0	54.4	49.9	42.3	26
75 and over	361.5	376.1	338.1	412.1	371.5	319.7	310.2	336.2	352.6	314.0	13

Statistical Bulletin, Metropolitan Life Insurance Company 57:p.5 Sept., 1976

Pneumonia as a general medical experience in our home is very common. It is more frequent during the winter months. We have seen very little of the characteristic influenzal pneumonitis (necrotizing tracheobronchitis) in recent decades. Our therapeutic results with pneumonia are variable but recovery is not uncommon although we have no statistics on rate and frequency. Our records do not show a great ponderance of terminal pneumonia in the winter season.

A review of the diseases associated with terminal pneumonia revealed a wide variety of pathologic conditions commonly seen in the aged, such as disability caused by old or, at times, recent hip fractures, cerebral disease, including hemiplegia with disability, chronic brain syndrome with nutritional and other physical problems, and heart failure in varying stages. Neoplasms were found in about 15% of our patients in diverse stages of dissemination. All of these pathologic processes are also seen at postmortem examination of patients without terminal pneumonia. Although aspiration as a cause of pneumonia is not uncommon in cerebral disease and other debilitated states and no doubt contributes to a higher incidence of terminal pneumonia, a large number of our patients expired with pneumonia without gross evidence of aspiration. We do occasionally see death due to aspiration of digested food and hence cannot dismiss the possibility of aspiration as a contributing cause.

The greater incidence of pneumonia with advancing years can be ascribed to many causes. The restrictive physiologic changes in the respiratory apparatus occurring in the aging process and deficits in the immunologic system are factors chiefly responsible for decreased pulmonary resistance to infection. Hence, it may well be anticipated that terminal pneumonia will continue to show a high incidence in the elderly. However, a reduction in the contribution of associated disease can lead to reduced mortality from terminal respiratory infection. Improved control of cardiac complications, correction of physical and neurologic disability and early and successful treatment of disabling injuries may well reduce terminal pneumonia to a minimum.

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Orthopaedic Care in the Institutionalized Elderly

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In dealing with the institutionalized elderly, we are dealing with a group of individuals that no longer can fend for itself. They become institutionalized because of medical, dietary and emotional problems; and they bring their problems with them. To the treating orthopaedist, they have essentially the same problems in similar proportions as the noninstitutionalized elderly. However, therapy must be tailored to their special status. They are sicker, and less able to cooperate, have a shorter life expectancy, and have a diminished need for perfection in anatomical restoration. What we aim for is pain relief, return to some useful function, and the least interference with their life patterns. Whenever possible, procedures should be definitive, one-stage affairs with the least possibility of complications. Walking with a painless limp is better than not walking at all. Even standing and taking one or two steps makes institutional care much easier. Their problems are many. Arthritis in all its forms, osteoporosis (symptomatic or not) with multiple vertebral collapse, and fractures, make up the largest set of problems we encounter, though the mundane problems of shoe fitting and its effects on mobility are not only time consuming, but are frequently insoluble. For example, an elastic usable shoe for a patient with intermittent edema associated with chronic congestive failure has yet to be adequately designed. If you combine this with severe hallux valgus and peripheral vascular disease (and an anxious daughter standing in the wings), you come to realize why the special groups of soul and body healers are people who have a "calling."

Elsewhere in this issue, there is an article on the medical treatment of arthritis in the elderly. In general, in the institutionalized elderly, surgical

therapy is not usually indicated. Their ability to cooperate in a rehabilitation program of any sort is very limited, and staging a series of procedures over a period of time exceeds their life expectancy. We do not do joint replacements on the residents of our institution.

Osteoporosis is another major problem. In its pure form, it is rare in this group. (It should be noted that the large majority of the residents in any geriatric nursing facility are women making up 90% or more of the population.) Its most frequent symptomatic area is the spinal column and it is accompanied by arthritis of every degree, and frequently with vertebral collapse—often spontaneous; but more frequently, it is associated with only minor trauma. Here, we are almost nihilistic in our approaches, using a simple back support, a calcium-Vitamin D preparation, analgesics, and rapid mobilization. On occasion, we utilize synthetic so called anabolic steroids, Dianabol or Durobolin. We have, in the past, used estrogen and testosterone combinations with varying success. Hormonal balance was a problem, and though we did seem to reduce the pain of osteoporosis—hirsutism, voice deepening, breast pain and vaginal bleeding all were seen. We have had symptomatic favorable response with anabolics without these side effects. As with all steroids, fluid retention, edema, and C.H.F. are ever possible consequences and these preparations must be used with extreme care. Daily weighing of patients is a relatively simple technique in helping in control. We have not used fluorides in institutionalized patients, though there are theoretical reasons why they may be indicated. We have not found it necessary in this population.

Up to this point, we have not dealt with life threatening conditions per se. However, with fractures, the story is different. Eliminating pathologic fractures that are a special problem, the most frequent fractures seen are Colles fractures of the

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wrist, surgical neck fractures of the humerus, and hip fractures. The epidemiology is alarming.

Wrist fractures respond, even without an anatomical reduction, to six weeks of immobilization. The wrist may not look pretty, but it works well: Shoulder fractures, similarly respond to simple slings, collar and cuffs, or any of a variety of shoulder immobilization devices. We have not operated, even on the four-part fracture variety. We do mobilize them early, within two weeks if possible. Our cases end up with painless shoulders even if their range of motion is not what it had been; but it does suffice for their needs. This is not to imply that all elderly patients should be treated this way. It must always be kept in mind that this is a population with limited needs, and though we strive for the best we can get, we must limit our goals to their realistic potentials. Fractures of the hip are a very special case.

It has long been known that fractures here, like Osler's pneumonia and pleurisy, are a precursor of death. Jacob (1) on his deathbed implored his son Joseph to steady his thigh (presumably to limit motion and attendant pain) while he gave them his parting thoughts. Bick (2), in his classic *Source Book of Orthopaedics*, quotes Astley Cooper's justified pessimism in regard to hip fractures. Cooper (3) was also the first to note the different morbidity and mortality in different varieties of hip fractures.

In terms of classification, we divide hip fractures into two primary groups: intracapsular and extracapsular. Within these two groups there are sub-classifications.

We can divide the intracapsular fractures into:

- a) Impacted fractures
- b) Base of neck, and essentially nondisplaced vertical fractures
- c) Displaced fractures

The treatment for all three types should be surgical, except in special circumstances.

Not requiring surgery are fractures in nonwalkers, the very elderly (90–95 years of age depending on other factors), when there is no pain, or when this fracture is noted accidentally on a film taken for another reason. Frequently, a displaced fracture becomes pain free in a very short time. The patients can then be mobilized, even walk with a walker or cane. The decision has to be made on the basis of the functional status, medical condition and discomfort the patient exhibits.

The majority of intracapsular hip fractures do undergo surgery. In displaced fractures, we routinely use a straight stem fenestrated Austin-Moore

(4) prosthesis, utilizing a head size that comes closest to the size of the head removed. Standard head sizes come in $\frac{1}{8}$ inch increments. When the rare occasion occurs that a size falls exactly between two sizes, we utilize the larger head size and have had no difficulties with it. The straight stem prosthesis functions by what is called an interference fit and would seem to have a theoretical advantage for mechanical distribution of forces over the curved stem prosthesis. I cannot really criticize the surgeons who employ a curved stem device, since the strain put on the device by this group of patients is not great, and the curved stem prosthesis is easier to insert.

For the very rare group of people who have a base of the neck fracture, or an undisplaced vertical fracture, we employ a Jewett nail that gives good fixation and a high degree of successful healing.

Impacted valgus fractures do well with any good fixation device. We have employed a variety of screw type appliances varying from the Lippmann (5, 6) screw bolt with its cork-screw shape down to multiple threaded Knowles pins. All seem to work well.

There is a special group of patients where we do a "Girdlestone" procedure. This is a rapid, pain-relieving technique wherein the femoral head is removed, any sharp bone edges of the neck trimmed, and the femur allowed to elevate to a stable position against the ilium. We use this instead of a prosthesis when the patient has had a colostomy since the infection rate with a prosthesis is very high. In the group of nonwalkers when pain is a prominent feature, this procedure is also used. It is, of course, also used as a salvage procedure when a prosthesis or other surgery has failed, either because of infection or for other reasons. Finally, we should mention that in using a prosthesis for a bone that is extremely porotic and almost pipe-like, a nonfenestrated (solid) prosthesis may be inserted that is bonded to the femur with methyl methacrylate. This is a rare occurrence—but it brings up the point that one must have a voluminous armamentarium before embarking on a surgical endeavor.

We do not attempt to reduce displaced intracapsular fractures with sliding devices of various sorts hoping for bony union. A prosthesis is definitive, essentially a one-time stress. The combination of nonunion, device failure, aseptic necrosis and surgical failure (infection, etc.) have a combined total that can run as high as 40% (7, 8). It is our feeling that this is far too high a price to pay.

The intertrochanteric fracture is a far different

problem. Here bleeding is more severe, morbidity and pain are very prominent, and the threat to life is greater. Classification is also complex. Simple undisplaced fractures usually heal well, rapidly, and the patient can anticipate restoration to a functional state in a few short months. Fixation is easy and blood loss not too severe.

At the other extreme the severely comminuted displaced fracture, with broad bleeding bone surfaces and massive blood loss, defying ready anatomical restoration and internal fixation, tries the Orthopaedist's ingenuity and patience. In general, these fractures will heal and heal well if we can create a stable fixation of the major bone fragments (the femoral neck, head and the femoral shaft) and hold it by an internal device. We prefer a Jewett nail, that is a canulated trifinned nail with a multi-holed plate device which transfixes the fracture. However, many other devices have been used with sliding screw-plate fixation mechanisms that allow coaptation of fragments becoming more popular at the moment.

In this, fracture surgery is almost always mandatory. These patients without surgery stand twice as much risk of dying as with surgery.

Tables I to V refer to a study done on 160 consecutive female patients at Kings County Hospital on two parallel services in 1951. On one service, all patients were operated on as soon as possible after admission. On the other service all patients were treated nonsurgically, by traction, casting, well-leg traction, or bed rest alone. The mortality figures refer to those who were alive 12 weeks after admission. The contrast of operative as compared to nonoperative results is astonishing. The original study was intended to include a far larger number of patients but was abandoned when the early fig-

TABLE I

160 Female Patients

MORTALITY 12 Week Survival

Alive	105 (66%)
Dead	55 (34%)

TABLE II

Acute Female Fractures

Total 160

Type	
Intertrochanteric	86 (54%)
Intracapsular	74 (46%)
Treatment	
Nonoperative	67 (42%)
Operative	93 (58%)

TABLE III

Intracapsular Fractures

Total 74

Treatment	
Nonoperative	31 (42%)
alive	18 (24%)
dead	13 (18%)
Operative	43 (58%)
alive	33 (45%)
dead	10 (13%)

TABLE IV

Intertrochanteric Fractures

Total 86

Treatment	
Nonoperative	36 (42%)
alive	15 (18%)
dead	21 (24%)
Operative	50 (58%)
alive	39 (45%)
dead	11 (13%)

TABLE V

Intertrochanteric Fractures		
Nonoperative	36	Died 21
Operative	50	11
Intracapsular Fractures		
Nonoperative	31	Died 13
Operative	43	10
Nonoperative mortality		51%
Operative mortality		22%

ures were tabulated and both services became "surgical."

The use of surgery as a therapy for hip fracture is fairly new, dating only to the 1930s with Dr. R. K. Lippmann (5, 6) being in the forefront of championing surgery and developing an early device, now infrequently used, for intracapsular hip fracture. Earlier procedures had been essentially reconstructive, though in the 1920s early attempts at blind pinnings had been attempted. The real thrust came after World War II and, though devices are still being designed and improved, the basic principle remains the same—restore the patient to function without pain and rapidly, with the least stress on the patient, and the least period of rehabilitation consistent with that function.

Technique

A word on patient care is in order. After many years, a standard care program for the patient has been arrived at. It is not perfect; it is not inviolate and can be modified as need arises. Surgery is done as soon as feasible after the patient has had a basic

check of the medical status. This is usually within 24–48 hours after hospitalization. If transfusions are needed they are given, usually as packed cells, to an adequate level. Blood is always available if needed in the operating room. On rare occasion Rh+ blood may have to be considered for Rh- patients. This has never posed a problem in our experience. Surgery is usually done under general endotracheal anesthesia. This facilitates pulmonary toilette at the end of the procedure, gives good respiratory control, is rapid, and reduces operating time. Whenever possible we try to limit our operating time to one hour. Anesthesia must be thought of as a controlled poison, and at this age the ability of the patient to detoxify these agents may be diminished. It is better to accept a stable fixation that is nonpenetrating, even if it is not optional, and have a viable patient than to extend a procedure inordinately for fear of a colleague's future criticism. That is not to say that one should do sloppy surgery, but duration of surgery has a definite relationship to infection rates and general morbidity.

We have not personally employed some newer devices, such as the Enders intramedullary nail. It undoubtedly has a place in treating some of these fractures but as yet, the radiation exposure, the technical difficulties of insertion, the backing out of the nails and the position of the extremities during the procedure seem to be technical obstacles that have not shown great enough advantages over other available devices. We catheterize all our patients before surgery and remove it as soon as we feel the patient is continent and able to cooperate. We think it reduces wetting of dressings and subsequent infection. At least in the immediate postoperative

period, this makes the patient more comfortable, and nursing care easier. We mobilize our patients the day after surgery, use compression bandaging on their calves, and protective devices to lessen pressure areas on heels and maleolii.

Summary

An overview of the orthopaedic problems encountered in a nursing home setting has been reviewed with a special emphasis given to the very real problem of the fractured hip. It is not exhaustive in any detail, and each item is worthy of far deeper consideration. It is addressed to the general physician, and even the nonphysician, with a few details and opinions that may be of interest to orthopaedists, a most individualistic lot.

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Origin and Evolution of the Walkerette

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Charles M. Williams was twelve years old when he went to his father's basement workshop, in the fall of 1924, to work on a walking aid for his Aunt Frances. He could hardly imagine then that his device would, in time, become the most widely used aid to the disabled, particularly the elderly, in the world. The story of how this evolved should be recorded.

Charles had observed that his aunt, Frances Ryan, recovering from a hip fracture at the Good Samaritan Hospital in Cincinnati, managed to slowly walk about in her room by standing behind and pushing the back of her armchair. The wooden legs then slid fairly easily on the wooden floor. She was too weak and unsteady to use her arms to walk with, as is basically required with crutches. Nor could she use the large framed "walker" on wheels that was the only other device generally available in those days. This was a most fearful machine, one that the elderly generally dreaded. When they tried to use it, they could not control the freely rotating wheels spinning on the vertical axes as well. In effect, the "walker" walked or, rather, careened and the patient held on for dear life. Many survivors of hip fractures either never walked again or took many months before they learned to get about. They couldn't use crutches or the walker on wheels. These patients simply lacked the transitional secure support between the wheelchair phase and independent walking.

One may speculate about why a simple, stable walker had never been designed before. There appears to be no previous record of a simple, four-legged device without wheels used as a walking aid.

This first walkerette designed by Charles was made of wood in essentially the same general shape as the millions of aluminum walkerettes now seen

everywhere in the world. It was simple and light; and it worked (Fig. 1).

Frances Ryan was immediately able to get about more freely. She soon developed appreciably more strength, stability, and confidence and was able to be discharged to her home taking her walking aid with her.

The hospital staff was most impressed with the potential of this new device. The superintendent, Sister Theodore, felt that additional walkers should be made for regular hospital use. She asked Elmer Frederic Reis, a design engineer for a hospital equipment company, to construct a heavy duty version. He modified Charles' design and constructed six walkers utilizing gas pipe three quarters of an inch in outside diameter.

The walking frame, as they then called it, was later duplicated some three to four hundred times in the next twenty years.



FIG. 1. The original wooden walking aid.

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One may, again, speculate as to why this successful device was only duplicated several hundred times in twenty years. We must recall that it was designed for the elderly with hip fractures. In the days before antibiotics and corrective surgery all too many of these unfortunate patients succumbed to hypostatic pneumonia in traction in geriatric wards.

It was not until 1945 and the development of sulfa drugs (sulfanilamide, sulfadiazine), penicillin, and more aggressive corrective surgery that the need for such a device suddenly broadened. Mr. Reis then modified the aid for commercial manufacture, constructing it with one-half inch steel conduit tubing with a cleaner design, lighter weight, and with a broader applicability (Fig. 2).

For approximately ten years, this was the only walkerette available and the basic design remained essentially unchanged.

In 1955, rubber tips capped with stainless steel glides were added to afford easier gliding with less vibration. This permitted less frequent lifting with each step as floor surfaces allowed. Also about this time, light weight adjustable-height aluminum walkerettes began to appear with standard rubber tips. As a result of further medical advances, the use of the walkerette began to expand tremen-

dously. In the next ten years, over a hundred manufacturers came into the field and distributed walkerettes by the hundreds of thousands in this country and overseas. Dozens of modifications were developed including models that folded, others that facilitated toileting, some that eased ascending and descending steps and others that provided folding seats. Accessories were added including elevated attachments for under-arm rests, to be used by patients with various disabilities, ashtrays clipped on, and various types of bags designed to hold personal items, food, and newspapers. Some models had optional wheels, others had brakes.

Unfortunately, 1970 saw the demise of the rubber insulated stainless steel glider tips, a secondary effect of the new OSHA Federal safety rules relating not to the device but to its method of manufacture. However, hundreds of thousands continue anew each year to be helped by the now standard rubber-tipped, light weight adjustable aluminum walkerette.

We, at the Jewish Home and Hospital for Aged, having learned to appreciate the value of the gliding walkerette, have been able to achieve a workable compromise that we feel has facilitated our management procedures. We simply attach chromium plated furniture glides to the rubber tips to afford a reasonable facsimile of the original. We find that the glider facilitates walking on our vinyl tile floors and, after 25 years of clinical experience, we feel that no resident who would be safe with a rubber tipped walkerette would be any less so with glider tips. Indeed, we have many residents who cannot walk with the rubber tipped walkerette but do so beautifully with glider tips.

Our fundamental approach is to attempt to retain or obtain as normal a gait pattern as possible. We find this more feasible with the gliding walkerette which does not require repeated "stop-elevate, drop-start" intermittencies so characteristic of rubber-tipped walking. It is difficult, oftentimes impossible, to learn new gait patterns when you have been walking with long established synchronous trunk and extremity rhythms for seventy-five or more years.

Organic Brain Syndrome (OBS)

During the past ten years we have been seeing increasing numbers of patients with varying degrees of OBS who also have instability, insecurity, at times, frank ataxia in standing and walking.

Many such patients during the earlier milder stages can use the gliding walkerette safely and



FIG. 2. The first commercial walkerette (1945).

reliably. They cannot readily learn an interrupted stop-start rubber-tip pattern and would, therefore, be limited to the wheelchair.

In later stages, they may require attendance and occasionally manual support even with glides. With rubber tips, many such patients cannot walk at all even with manual support.

The difference in ease and freedom offered by the glides is deeply appreciated both by the patients and the nursing staff.

Status Post-Hip Fracture

Newer surgical techniques following hip fracture are dramatically shortening postoperative courses. With the gliding walkerette we can progress quite satisfactorily during initial non, minimal or partial weight-bearing stages. Our patients find it easier to push the glider forward than to pick it up and thrust it forward with each step. Once progressive weight bearing is permitted, patients return more rapidly and confidently to a smoother normal gait with glides without the interrupted sometimes bizarre and noisy intermittencies of newly learned rubber-tip walking.

Status Pre-Hip Fracture

We have believed for years that many patients with hip fracture were truly disabled before the injury. Detailed histories all too often revealed descriptions of varying degrees of erratic instability, loss of balance, near-miss falls, leaning on walls or furniture and the use of one or two canes prior to the ultimate fracture. It has long been established that all requests for canes be referred immediately to our service. In many instances we feel we have been able to permit safe, independent walking with gliding walkerettes for years with residents who are, otherwise, potential candidates for early fracture. Indeed, we feel that we are preventing more fractures than we are treating.

Such procedures, incidentally, are also valid in

community practice. The broadloom carpet permits the glider almost as well as the kitchen and the bathroom floors.

Status Post-Cerebrovascular Accident with Moderate Hemiparesis

The use of the gliding walkerette permits bilateral support while re-establishing a smooth reciprocal gait pattern.

When the hemiplegic extremity is too paretic for functional grasp, we put glides on a unilateral quadriparted "walkane," to be held by the good hand, and permit practice toward normal rhythms rather than complicate with intermittent intricacies.

Convalescents Following Acute Medical Illness: Myocardial Infarctions, Congestive Heart Failure Episodes, Acute Chronic Obstructive Pulmonary Disease Flare-ups, Pneumonia. Complications following Surgical Procedures: Pacemaker Implantation, Gastrointestinal Tract, Genitourinary Surgery

Genitourinary Surgery

The elderly convalescent requires cautious, graduated mobilization in normal everyday patterns including walking. Intermittent weight lifting while disrupting a normal gait pattern is not necessary with the gliding walkerette.

Amputees

We routinely have amputees with prostheses, above or below knee, learn to ambulate quite freely with the gliding walkerette, adeptly joining lunch hour and recreation room traffic jams.

The ultimate walkerette has yet to be developed with flexibility for all surfaces. In the meantime, those we now have will simply have to be lifted when necessary.

And we remain in debt to the young Charles and his simple idea to help Aunt Frances.

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Rheumatic Disease in the Elderly

SELVAN DAVISON, M.D., F.A.C.P.

The care of rheumatic conditions (arthritis and allied disorders) in all age groups and, certainly in the elderly, depends primarily upon a correct diagnosis (1). Too often, pain syndromes, particularly in older people, are dismissed as "it's your age," and/or "you have arthritis, learn to live with it." But "all that pains is not arthritis" (2). Neoplasms and leukemias, the bone lesions of multiple myeloma, amyloid deposition producing neuropathy, diabetic neuropathy, peripheral vascular diseases such as Leriche syndrome, and other local and systemic diseases may produce pain and disability needing to be differentiated from specific rheumatic conditions. Once a correct diagnosis of arthritis is established, a proper course of therapy can be followed, often producing a significant amelioration of physical symptoms and frequently removing the spectre of crippling disability.

A complete description of the rheumatoid diseases can be found in standard textbooks of rheumatology. This article will present some of the important, more common, and significant features of rheumatic disease in the elderly. Such syndromes as Takayasu's, Reiter's, the arthropathies associated with intestinal disease, and ankylosing spondylitis are diseases of the young and will not be discussed, although the burnt out stage of the latter is of course found in older men.

Rheumatoid Arthritis

This is a systemic disease with a distinct inflammatory process in the involved joints. While it is certainly more common by far in the younger age group, it is not at all unusual in the elderly (3-5). It may be seen as the chronic form or a continually active form over many years; it may also be seen as an acute disease beginning in the older years. This latter entity may differ somewhat from the classical

rheumatoid arthritis of the younger patients. While in the younger age group, there is a significantly higher ratio of female to male, generally 3-4:1, in the elderly the rate decreases almost to 1:1. Onset in the elderly is often more explosive, yet may run a shorter course without occurrence of significant deformity. However, about 75% of rheumatoid arthritis in the elderly is not noticeably different in course and prognosis (6). Erosions, rheumatoid nodules and arteritis are uncommon.

TREATMENT: The treatment in the elderly is much the same. It includes the use of nonsteroidal antiinflammatory drugs, gold, penicillamine and antimalarials; and there is no difference in side effects or results. In the elderly, proper employment and interrelation of a rehabilitation program and social services is vital.

Corticosteroids play an important role, but their indiscriminate use is to be condemned. Particularly in the elderly, high-dose steroids often produce devastating ill effects, such as vertebral collapse (osteoporosis), enhancement of diabetes mellitus, psychoses, hypertension, peptic ulcer and suppression of the adrenopituitary axis. These undesirable reactions far outweigh the benefit. However, parenthetically, when other methods have failed, sufficient relief and return to a liveable life style can be achieved with small doses, 10 mgm or less daily, of prednisone. In this manner, the serious dangers are almost entirely avoided.

Modern reconstructive surgery has greatly altered the outlook when there is severe deformity and loss of function. Such surgical procedures, particularly total joint replacement have produced a major advance and is a rational approach even in the eighth and ninth decades.

Osteoarthrosis (Osteoarthritis, Degenerative Joint Disease)

The etiology of osteoarthrosis is not clear and is not monovalent. While it is a common finding in

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advancing age, other factors must be considered. Previous trauma, congenital conditions (for example, congenital hip dislocation), occupation, weight, sex and metabolic disorders may contribute. Nevertheless, a distinct correlation exists between osteoarthritis and age (7). But a very significant distinction must be made. While as many as 85% of people over 55 years of age will show x-ray evidence of osteoarthritis, only between 15%–20% will experience symptoms (8, 9). The asymptomatic patients with x-ray evidence of arthrosis should not be told they have arthritis with resultant engendering of the fear of becoming crippled.

The diagnosis generally is not difficult. Most commonly affected are the knees, hips, terminal interphalangeal joints and the first carpometacarpal joints. When there is pain, stiffness, and limitation of motion, this is usually without signs of inflammation. However, in the knee joint, swelling and warmth are not too rare, but the synovial effusion is not inflammatory.

Heberden's nodes that affect the terminal interphalangeal joints are quite well known and easily

recognized. Less well known, but far from uncommon, are Bouchard's nodes that affect the proximal interphalangeal joints and too often cause an incorrect diagnosis of rheumatoid arthritis. These nodes are characteristic, being asymmetrical in appearance and distribution, do not involve the metacarpophalangeal joints and have a distinct x-ray picture of narrowed joint space, osteophytes and sclerosis (Figs. 1, 2). In some very severe cases of osteoarthritis, erosions are present and must be differentiated from rheumatoid arthritis (10).

Spondyloarthritis

Osteoarthritis in the spine, despite extensive x-ray changes, usually does not produce pain. As in any age group, low back and cervical syndromes are mostly caused by disc degeneration and not osteophytes. Cervical neuropathies and myelopathy often follow degenerative changes in the apophyseal and neurocentral joints, and development of a bony transverse ridge.

Osteophytic narrowing of the intervertebral fo-



FIG. 1. Rheumatoid Arthritis. Note the severe juxtaarticular osteoporosis and the uniform proximal interphalangeal and metacarpal phalangeal joint involvement.



FIG. 2. Osteoarthritis. Note that despite the significant erosions, there is terminal interphalangeal joint involvement and asymmetric proximal interphalangeal pathology.

ramina is an important problem in the elderly. This narrowing causes compression of the vertebro-basilar artery which courses through the foramina, and consequent cerebral ischemia.

In the elderly, disc degeneration of L4-5 and L5-S1 is much less common. More often the upper lumbar discs are at fault and nerve root compression here produces radiation to the crural areas instead of the usual sciatic distribution to the lower extremity.

Diffuse Idiopathic Skeletal Hyperostosis (Forestier's Disease)

This is a disorder affecting approximately 8% of the elderly population (11). It appears mostly in the spine and x-ray reveals continuous ossification along the anterior aspect of contiguous vertebrae. Despite slowly progressive and significant pathology, many patients are asymptomatic. Stiffness and backache may develop but usually are not very severe.

Avascular Necrosis (Aseptic Necrosis)

This is a syndrome of varied etiology (Lupus erythematosus, steroid induced, Caisson disease, sickle cell, among others.) An important cause in the elderly occurs following nailing of a hip fracture. Because of the trauma and/or the metal fixation, the vascular supply to the femoral head is compromised and the patient complains of increasing pain and disability. Medical therapy will be without value and x-rays will reveal the avascular pathology. Treatment is removal of the involved head and neck of the femur and a total hip replacement.

Treatment of Osteoarthritis

- 1) Use nonsteroidal antiinflammatory drugs.
- 2) Avoid systemic corticosteroids as being valueless in osteoporosis.
- 3) Employ intra-articular instillation of corticosteroids.
- 4) Use physical therapy and rehabilitation. Traction in the elderly generally is not very effective and may be dangerous, particularly cervical. Judicious use of heat modalities, gentle exercise, cervical collar and lumbosacral corsets are generally helpful.
- 5) Orthopedic surgery, particularly total hip replacement, has been of inestimable value.

Polymyalgia Rheumatica and Giant Cell Arteritis

These two conditions are peculiar to the older patient and do not occur under 55 years of age and almost always over 60 years of age. Polymyalgia Rheumatica has an acute onset, sometimes overnight, of severe pain and stiffness of the pectoral and often pelvic girdle as well; usually with tender muscles but without atrophy or true weakness. Low grade fever and anemia occur. The only specific laboratory finding is a markedly elevated sedimentation rate, often over 80 mm/hr Westergren. There is no arthritis, the rheumatoid factor is negative and prednisone dosage of 10 mgm daily produces rapid relief of symptoms and reduction in the erythrocyte sedimentation rate (12).

Giant Cell Arteritis (cranial arteritis, temporal arteritis) has a predilection for the same age group as polymyalgia rheumatica. Usually it involves medium-sized arteries with production of diverse symptoms. Most frequently, there is temporal artery thickening, tortuosity and tenderness. The danger is the frequent simultaneous ophthalmic artery pathology and this can and does produce

rapid visual impairment, often total blindness. Therefore, one must not wait for a temporal artery biopsy but when temporal arteritis is suspected, prompt initiation of treatment is vital with large doses (usually 60 mgm daily) of prednisone. This dose is to be tapered as indicated by the course of the patient, to prevent the ravages of long-term, high-dose steroid (13).

Many investigators consider Polymyalgia Rheumatica to be but a part of the giant cell arteritis syndrome. In some instances of polymyalgia rheumatica without clinical temporal artery involvement, a positive giant cell arteritis has been found on biopsy. However, in the personal experience of Dr. H. Spiera and myself with over 500 cases, we have found only three instances beginning as polymyalgia rheumatica eventually developing temporal arteritis. We have performed routine temporal artery biopsies in a number of cases of polymyalgia rheumatica and found no evidence of arteritis. We believe that in Polymyalgia Rheumatica without clinical evidence of temporal arteritis, high dose corticosteroid is not indicated.

Crystal Disease (Urate Gout)

Long ago, Hippocrates noted that gout in women was rare before the menopause. So that under the age of 60 years, 95% of gout occurs in males and, after 60 years, the incidence in females increases to 30%.

The definitive diagnosis in all age groups depends not only on the uric acid blood level that, even in the face of an acute episode, may be normal. It depends on exhibition of strongly negative birefringent monosodium urate monohydrate crystals in the synovial fluid.

Generally, gout in the elderly presents only a few significant differences.

- 1) Increased female incidence.
- 2) A reduced incidence of familial and socioeconomic factors.
- 3) An increased incidence of secondary gout (related particularly to underlying blood dyscrasias.)

The occurrence of tophi, x-ray changes, podagra, etc. is essentially similar.

Hyperuricemia itself does not necessarily indicate gout. Particularly in the elderly, diuretic agents are in frequent use and may elevate the blood uric acid level but usually without clinical gout. (A new diuretic, Ticrynafen, has a uricosuric action and will be useful when hyperuricemia exists and a diuretic needed.)

Treatment of Urate Gout

In all acute gout, the mainstays are colchicine, indomethacin and phenylbutazone. Colchicine in doses of 0.6 mgm once or twice daily may be necessary in long-term maintenance. Over the long haul, strict dietary restrictions except for truly high purine foods, are not necessary since the advent of uric-acid-lowering drugs. Probenecid inhibits reabsorption of uric acid; and allopurinol, a xanthine oxidase inhibitor reduces uric acid formation. When uric acid is over produced, and when there is renal disease, allopurinol is preferred. When large tophi are present, hydration and alkalinization of renal calculi is necessary.

In treating an acute attack, indomethacin or phenylbutazone will abort the episode. Indomethacin is given in a dose of 100–150 mgm the first day, in four divided doses, followed by 75–100 mgm a day for 3–4 days. Phenylbutazone, the first day, is in three doses of 200 mgms each, followed by 3–4 days of 100 mgms three times daily. Colchicine, 0.6 mgm b.i.d., is given simultaneously to prevent a rebound effect, and should be continued. If after a year no further acute attacks have appeared, Colchicine may be discontinued.

Chondrocalcinosis (Pyrophosphate Arthropathy, Pseudogout)

This is a crystal deposit syndrome found particularly in older persons (14). The sex ratio favors males by 1.5 to 1. It usually attacks the large joints, most often the knee. By x-ray, suspicion is aroused by noting linear calcification in the knee menisci, the symphysis pubis, or in the joint capsule as in the hip. The definitive diagnostic findings are weakly positive birefringent crystals of calcium pyrophosphate dihydrate in the synovial fluid. Although chondrocalcinosis may have an x-ray incidence as high as 27.6% (15), there is no correlation between the x-ray and the clinical occurrence of an inflamed joint.

In the presence of chondrocalcinosis, particularly when diffuse, one must rule out hyperparathyroidism and hemochromatosis. Also, an increasing number of rarer crystal-induced arthropathies are being reported and require identification.

Treatment of pseudogout is often not very rewarding. Usual antigout measures frequently are not very effective, but indomethacin and phenylbutazone should be tried. Aspiration of the joint with or without instillation of corticosteroid is most helpful. Avoidance of trauma is the best method of

avoiding acute episodes but spontaneous attacks are common.

Soft Tissue Lesions

Shoulder-Hand Syndrome. This entity was named by Steinbrocker (17). It represents a reflex dystrophy and is most common in older patients, and often follows a myocardial infarction, a cerebrovascular accident, trauma, cervical discogenic disease, and pericapsulitis or peritendonitis of the shoulder. About 15%–20% of cases are idiopathic. Treatment with corticosteroids systemically, or stellate ganglion blocks, is often effective. Tendonitis, capsulitis, myositis, epicondylitis, painful heel (with or without a spur), various bursitides and entrapment syndromes are generally similar in all age groups as to clinical appearance and treatment.

Systemic Connective Tissue Disorders

Rheumatoid arthritis has been discussed. As with rheumatoid disease, systemic lupus, scleroderma, dermatomyositis, and polymyositis, Sjogren's syndrome and polyarteritis nodosa, while more prevalent in the young, do occur in the elderly. (When polymyositis, particularly in males, occurs over the age 40 years, a search for an occult neoplasm is mandatory.) Most important is that these diseases are usually more insidious in onset but usually run a more benign course in the older than the younger age group.

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Common Skin Problems in an Aged Population

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The incidence of significant skin disease increases with aging. By age 65, 650 out of every 1000 individuals will have cutaneous problems (1). During a period of eight months (March to October 1979), approximately 300 patients among the residents at the Jewish Home and Hospital-Kingsbridge Center were referred to me for dermatologic consultation. The incidence of various dermatologic entities among these patients is reported in the following chart.

Chronic eczematous dermatitis	86
including stasis dermatitis	16
contact dermatitis	8
Seborrheic dermatitis	29
Seborrheic keratoses	26
Actinic keratoses	23
Psoriasis	23
scalp	7
body	16
Intertrigo	22
Basal cell carcinoma	18
Nonspecific pruritus without skin pathology	8
Skin tags	7
Sebaceous cysts	7
Tinea infections, all locations	5
Leg ulcers	5

The following conditions were seen fewer than five times: Acute eczematous dermatitis, cheilitis, milia, cutaneous horn, sebaceous hyperplasia, squamous cell carcinoma, prurigo nodularis, onycho-

dystrophy (non fungal), drug eruption, furuncle, impetigo, mucous cyst, insect bites, angioma, herpes zoster, hypertrichosis, radiodermatitis, cacinosis cutis, perleche, telangiectases, and verruca vulgaris.

The most common problems seen at Kingsbridge Center merit some discussion:

The greatest number of patients had nonspecific chronic eczematous dermatitis. Their skins were erythematous, lichenified and fissured, frequently with excoriations (secondary to pruritus). The backs of the hands, the forearms, shoulders and legs were most often affected. In about 20% of these patients, the etiology was relatively clear (venous stasis or contact dermatitis). All had dry skin; undoubtedly a precipitating factor in their dermatitis. Histologically, aged skin shows a thinned epidermis with a normal stratum corneum, shrinkage of the sebaceous glands and a decrease in the number and size of sweat glands (2-3).

The capacity to restrict water loss is normal, but it is thought that water from the dermis is inhibited from reaching the stratum corneum (4). The lack of moisture in the skin leads to excitability of the cutaneous nerves and thence to itching. Dryness is further enhanced by low relative humidity, overzealous bathing and strong soaps.

These patients can be helped immeasurably by a bedroom humidifier in winter and by restricted bathing. The application of emollient creams to the damp skin will also aid water retention. Topical steroids creams will diminish itching and lichenification.

Seborrheic dermatitis also occurs commonly in the elderly. This disease, of unknown etiology, causes "dandruff" of the scalp, and erythema and scaling of the forehead, nasolabial, post-auricular and anterior chest areas. Topical steroid creams with 2%-3% sulfur are effective therapy. Recur-

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rence, however, is common. What looks like seborrhea in some elderly patients, is actually accumulated scale from infrequent shampooing.

Seborrheic keratoses, mostly seen after age 50, are not related to seborrheic dermatitis. There appears to be a genetic predisposition to their formation; sun exposure is not implicated. They vary in color from black to brown to grey, have well-defined borders and a stuck-on appearance. Except for cosmetic considerations, they can be left alone. Removal by electro-desiccation and curettage however, is relatively simple.

Solar keratoses do occur in sun-exposed areas. Having the potential to form both basal and squamous cell carcinomas, they require biopsy and removal: either by surgical methods or with the topical application of 5 Fluoro-uracil cream (1-5%). Applied for two to three weeks, the 5 fluoro-uracil selectively destroys the actinic cells. In the process, a severe eczematous reaction temporarily occurs. The patient must be forewarned.

Psoriasis of the scalp is seen with thick, adherent crusts, usually in a patchy, rather than generalized distribution. The occiput is a common site.

Psoriatic lesions of both body and scalp are more often itchy in the geriatric patient than in younger sufferers. This is probably related to the general dryness mentioned previously. Some relief from scaling and itching may be obtained with topical steroids, but complete resolution remains elusive.

Intertrigo is seen most commonly in women with pendulous breasts. Shiny red areas occur with fine scaly borders. Exudation from these lesions may occur. When satellite pustules are evident, a secondary *Candida albicans* infection is present. One must be careful not to further macerate the area with ointments. Instead, cool compresses, steroid lotions and antimonilial powders, when indicated, lead to rapid resolution. Erythema under the breasts or in the groin also may be a manifestation of seborrhea or psoriasis. These patients generally have disease at other sites as well.

Basal cell carcinoma in the aged skin occurs, especially in fair-skinned individuals with a history of prolonged sun exposure. Limited, but intense solar exposure can also be causative (5). Therapy in elderly patients must be individualized. Small lesions can be surgically excised or electro-desiccated with ease. Radiotherapy for larger lesions often will be easier for the older patient to tolerate.

Generalized pruritus with a normal skin can be enormously frustrating for the patient and the doctor. Usually, hepatic pathology, diabetes, thyroid disease, polycythemia and neoplasia are ruled out. The etiology remains unknown and therapy unsuccessful.

Skin tags (acrochordae) of the neck, axillae and groin are bothersome. They itch, get caught on jewelry, and become irritated and infected from friction. They can be destroyed by electro-desiccation. The desiccated tag will fall off spontaneously in five to seven days.

Sebaceous cysts do not require surgical excision unless they are subject to trauma and infection.

The incidence of tinea pedis and onychomycosis was less in my series than generally reported in an aged population. Usually these infections are mild and asymptomatic and not referred to clinic for therapy. *Candida albicans* infections and erythrasma can mimic tinea of the tow webs and must be ruled out.

Leg ulcers at times defy all therapeutic attempts because of the underlying vascular pathology. Topically applied benzyl peroxide (5-10%) has shown great promise in healing many of these recalcitrant ulcers. The old fashioned Unna boot also can be useful. It gives vascular support and protects the ulcer from external and self-induced trauma.

The inexorable changes of aging produce a multitude of dermatologic problems. Many can be controlled, some quite easily with simple therapy, affording great comfort to the elderly patient.

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The Management of Cerebral Disorders in the Geriatric Population: A Pragmatic Approach

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Introduction

It is well known that the relative incidence of neurologic disease rises rapidly in the geriatric population. The patients are often labelled as suffering from stroke or hardening of the arteries when actually there is progressive diminution of mental functioning. Brief episodes of neurologic dysfunction such as weakness of one side of the body or confusional states are almost automatically given the label of "TIA" or "CVA."

To the neurologist, these labels are undesirable since they imply, usually unjustifiably, knowledge of both localization and pathogenesis. Clinical experience demonstrates that many different etiologic circumstances can result in similar patterns of neurologic dysfunction (1). The rule-of-thumb that vascular pathology usually starts suddenly but mass lesions start gradually is not a reliable guide. An awareness of the dynamic complexity of cerebral dysfunction in the aged is essential for proper diagnostic management and therapeutic intervention (2).

The neurologist is rarely the primary care physician. Therefore, the family doctor and internist must maintain a high level of sensitivity to evidence of the possible existence of cerebral lesions or dysfunctions. Since so many systemic diseases common to the elderly have cerebral manifestations, the need for this sensitivity is particularly great in dealing with the aged. Hypertension, diabetes mellitus, and carcinoma, as examples, are not only frequent causes of brain disease, but may often be first manifested by brain dysfunction. This is also

true of systemic infections, blood dyscrasias, renal failure, vasculitis, and other conditions. It is accordingly appropriate, especially in dealing with the geriatric population, that a close working relationship must be maintained between internist and neurologist. Even those patients who seem to have the most clearcut evidence of simple cerebral lesions should be fully screened by the internist either before being referred, or while under the care of the neurologist.

When a strong presumption of cerebral pathology has been established, an extensive array of laboratory methods is available for help in arriving at a specific diagnosis (3). The determination of which test is to be used in each case requires a high degree of sophistication and judgment. This is particularly so since a shotgun approach to screening tests can result in needless discomfort and even risk to the patient, as well as needless expense both to the patient and to society.

One example of a time honored but relatively valueless test is the skull x-ray. While on rare occasions abnormal calcifications or metastatic bone lesions may be found, plain x-rays hardly ever present distinctive diagnostic information in patients with chronic complaints. Although x-rays are essential to determine the presence of fractures, even in head injury cases, they are as likely to be misleading as they are helpful. An asymptomatic patient with a small linear fracture of the occipital bone may be rushed into the hospital with great fanfare while a patient with a negative skull x-ray and a subacute subdural hematoma may be sent home to quietly slide into coma.

The electroencephalogram (EEG) is a procedure that is wasteful if used as a screening procedure in an unselected population (4). It is, however, of great, at times even unique value in selected cases. The

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procedure reflects certain aspects of brain function, and, therefore, provides information that cannot be obtained by tests focussed on depicting physical characteristics of tissues and vessels.

Patients with mental changes, or paroxysmal cerebral phenomena of any kind, may be shown to have focal or generalized EEG abnormalities. Certain fairly characteristic EEG findings may indicate the presence of underlying metabolic disorder. The demonstration of an unequivocal EEG focus in a clinically paradoxical location almost invariably indicates the presence of a multifocal disease process (5).

An important specific application of EEG in the geriatric age group is in the nonsurgical management of brain tumor and subdural hematoma. Particularly in the latter case, the EEG is the only reliable laboratory method that can indicate the presence of progression which may necessitate surgical intervention (6).

It must be emphasized that the EEG is of benefit when used selectively, in the context of a problem formulated on the basis of clinical considerations. Nonspecific abnormalities are quite common in the elderly, and completely normal records may be seen in patients with severe Alzheimer's disease or even benign tumors (7). In ambulatory patients, particularly those with common nonspecific complaints such as dizziness, faintness, and disturbed gait, the EEG is rarely of value, and may even be misleading as a source of false reassurance (if normal), or unjustified alarm (if nonspecifically abnormal).

In view of the prevalence of occlusive vascular disease in the aged, and the increasingly active interest in its treatment by surgical means, techniques for noninvasive assessment of vascular function have multiplied in recent years (8, 9).

Direct and indirect methods of measuring retinal artery pressure (ophthalmodynamometry) are useful in helping to determine the patency of the carotid arteries. The efficacy of this test is enhanced when it is used in conjunction with sonographic methods indicating the direction of blood flow in the internal and external carotid artery circulation and the wave forms of carotid pulsations (Doppler sonography) (10, 11).

These techniques require considerable skill and experience in order for reliable results to be obtained. It is for this reason that they are not yet widely available or fully dependable. Another major drawback is a methodologic one. The ulcerated atherosclerotic plaque in the carotid artery, giving rise to platelet emboli, is considered to be one of

the most important surgically remediable lesions. These plaques cannot be detected by Doppler sonography. Angiography must be done to reveal such a lesion. Even with Doppler studies indicating a stenotic lesion of the carotid, confirmatory angiography must be performed prior to consideration of surgery. Since angiography is a procedure that carries a significant risk in this age group, especially in those for whom it is indicated, that is, those with lesions, it cannot be advocated casually (12). This is particularly true since reliable predictions of the long-term benefits of surgery for carotid stenosis cannot be made. Multifocal occlusive disease is very common in both brain and heart in the elderly, making prognosis very difficult. Newly developed microsurgical revascularization procedures have also been advocated but are at present completely unverified as a potential value (13).

Brain scanning with the use of radioactive isotopes is another noninvasive procedure that can be of great help in selected cases, but which is often misused as a screening test.

In dynamic brain scanning, a bolus of isotope is injected intravenously, and gross flow patterns in the neck and head are depicted during the seconds after the isotope reaches, and is pumped out of the heart. While not indicating patency of specific vessels (except perhaps the common carotid) this technique can give valuable information regarding vascular perfusion in brain regions affected by occlusive disease.

The static brain scan is a recording of uptake of the isotope as measured 45 minutes to three hours after injection. Highly vascularized lesions, or those affecting the permeability of the blood-brain barrier tend to show accumulation of isotope. Infarctions (between the fifth and 30th days) and metastatic tumors are particularly well shown. Subdural hematomas are shown with a high degree of reliability, a fact which is of particular importance in view of the failure of some of these lesions (isodense hematomas) to appear in CT studies of the brain.

As in the case of the EEG, the nuclear scan shows a prohibitively high cost-benefit ratio as a screening test in ambulatory patients with minor or nonspecific complaints. When the neurologist suspects serious pathology, however, this test can be of great value as a noninvasive method of gaining information which helps in deciding whether to hospitalize a patient for studies such as angiography.

Radioactive isotopes are also used in the detection of abnormalities in the pattern of flow and absorption of cerebrospinal fluid. While consider-

able controversy exists as to the diagnostic criteria for, and the proper management of, "normal-pressure hydrocephalus," such a clinicopathologic entity may occasionally exist. In this condition, the outflow of CSF from the ventricles, and its subsequent absorption is impeded. Ventricular enlargement and disturbed brain function result, causing various central nervous system (CNS) dysfunctions, as well as the classical triad of organic mental syndrome, impaired gait, and urinary incontinence. The abnormal flow patterns can be confirmed by nuclear scanning performed at 24, 48, and 72-hour intervals after the intrathecal injection of radioactive isotope.

The most important new, and truly revolutionary development in the management of patients with cerebral disorders is computerized transaxial tomography (CT scanning). The device uses finely collimated beams of x-rays from multiple rotating or stationary sources. Corresponding detectors are arrayed in such a way as to count the photons passing through intervening tissues. The computer analyzes the information received to give absorption values for blocks of tissue of predetermined size in each "slice" of the structure under study. These numerical data are then converted into pictures analogous to the x-ray photographs familiar to all physicians, in which the most dense structure such as bone appear light, and the least dense appear dark.

This apparatus makes it possible to visualize details of brain structure, without risk, in a completely unprecedented way. Even so, however, its use in clinical situations requires discrimination, and the results must be interpreted with discretion. The utility of CT scanning is greatly enhanced by the intravenous injection of iodide-containing contrast substances which intensify the differences in radiodensity between various structures, mainly by increasing absorption by highly vascular ones, or ones in which the blood-brain barrier has been breached. This procedure is not as innocent as simply CT scanning which is without risk, since it carries the risk of a potentially serious allergic reaction. Even so, however, it can be accomplished on an outpatient basis and is of great value in determining which patient must be hospitalized for more invasive diagnostic procedures.

Diffuse or focal atrophy, single or multiple neoplastic masses, infarctions, intracerebral hemorrhages, and subdural hematomas may be readily diagnosed by experienced neuroradiologists using this technique. Hospitalization is often necessary

for angiographic verification of potentially remediable lesions. Even in the elderly, benign tumors in some locations may be successfully removed with good long-term results. On the other hand, CT scanning makes possible periodic noninvasive monitoring of an angiographically verified benign tumor in cases in which nonsurgical treatment has, for one reason or another, been decided upon. Malignant tumors may be irradiated, or treated with chemicals.

The apparent easy accessibility of diagnostic information by CT can be misleading. The need for proper correlation of the total clinical situation with these data cannot be overstressed. It is not simply parochial self-interest that impels us to caution against the tendency for internists to bypass the neurologist by relying on the results of laboratory tests, particularly the CT scan. Specialized training and experience is necessary for a balanced approach to the "what?, when?, and where?" of testing in clinical management. It is important at all times, particularly in dealing with geriatric patients.

The discomfort, potential disorientation, and actual danger of iatrogenic illness makes hospitalization something to be avoided whenever possible. Catastrophic events may make hospital care unavoidable. When the history is relatively slow and progressive, however, it is frequently possible and always preferable to handle the patient on an ambulatory basis. It is in doing so that the clinical judgment of the neurologist is crucial. Careful clinical observation during the evolution or resolution of the illness is a tool that is often undervalued. It may, however, be the most useful one, even when used in conjunction with carefully chosen laboratory methods.

The following cases illustrate some of the points that have been made.

Case #1. The patient is a 73-year-old woman who suddenly became ill. Her son telephoned her in the morning and did not obtain an answer. He immediately went to his mother's apartment and found her "distant and perplexed in the dining-room." Also "the right side of her body seemed weak." She was seen by her family physician, and as her symptoms began to diminish rapidly he decided to request neurologic consultation on an elective rather than emergency basis. Improvement had continued by the time she was first seen by a neurologist, five days after the acute onset. The neurologic examination at that time revealed an alert lady who was apparently aphasic, and had a mild but definite hemimotor and hemisensory syndrome. An electro-

encephalogram was performed, and revealed 1 to 2 cps activity, up to 150 microvolts, over the left cerebral hemisphere. There was no single focus, but there was predominance of the slow-wave activity over the temporal lobe.

A CT scan was performed, and revealed "a relatively ill-defined area of faintly increased density in the left frontal region which is partially surrounded by a small zone of lucency. With contrast enhancement a fairly discreet rim of increased density became obvious and outlined the lucent zone surrounding the faintly dense mass lesion." The radiologist's impression was that this was "probably a resolving hematoma—the possibility of a neoplastic process cannot be totally ruled out" (Fig. 1).

Because the patient was continuing to improve, the decision was made to continue observation on an out-patient basis. A repeat CT scan one month later revealed that the size of the lesion had decreased considerably, and the final diagnosis was "resolving intracerebral hematoma, left frontal region" (Fig. 2).

Case #1 well illustrates the situation in which a patient, even with fairly alarming manifestations of localized cerebral dysfunction, can properly be managed at home with diagnostic laboratory procedures carried out on an ambulatory basis.

Case #2. The patient is a 76-year-old diabetic, hypertensive woman who was admitted to the hospital by the neurologist, sight unseen, after having been informed by the family physician that she had a 48-hour history of focal seizures involving the left hand that were increasing in duration, and occur-

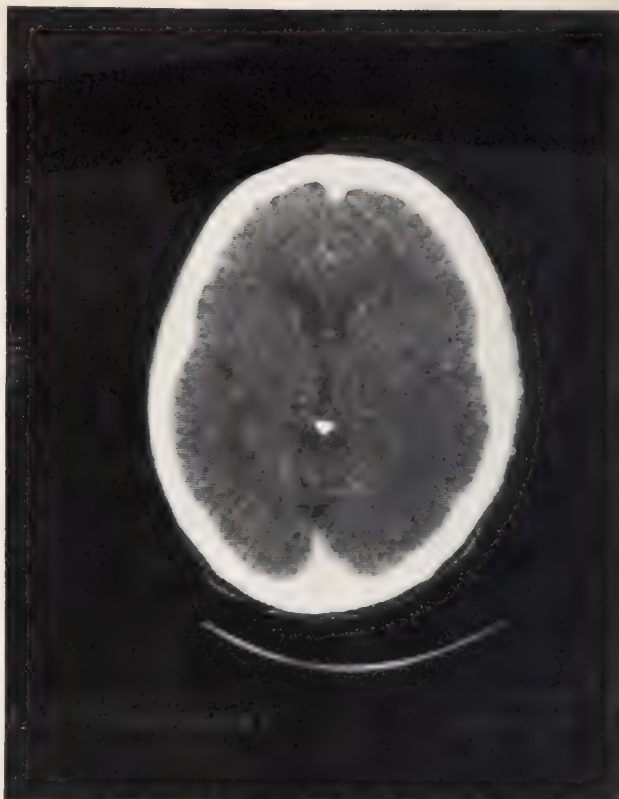


FIG. 2. Resolving left frontal lesion.

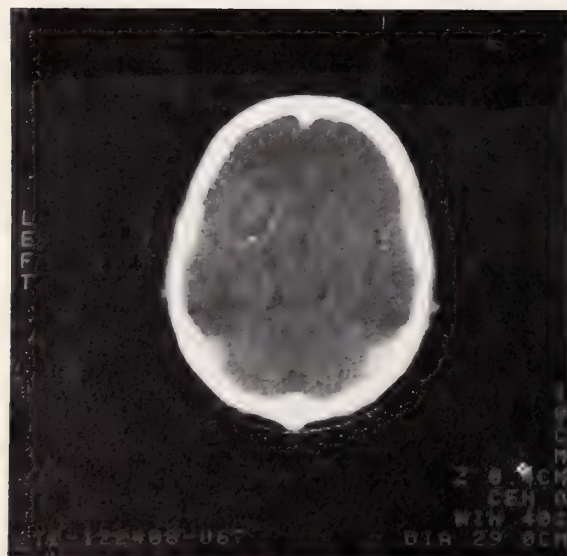


FIG. 1. Left frontal lesion.

ring at shorter and shorter intervals. On admission to the hospital, she was alert and oriented, showed a normal mental status, and had a completely negative neurologic past history. She had almost continuous clonic twitching of the left hand and, at times, showed movements at the elbow and shoulder as well. During the interval between the convulsive episodes, there was marked weakness of the left upper extremity. She was treated with large doses of intravenous phenobarbital and diphenylhydantoin, but even so the seizures persisted for 36 hours. An electroencephalogram performed while the seizures were present revealed no definite abnormality. Isotope brain scan showed a zone of increased density in the right parietal region, and CT scan revealed a large radiodense mass in the right parietal parasagittal region. Angiography was performed, and a vascularized lesion with the radiographic characteristics of meningioma was found to be present. Four days later, surgery was performed and the tumor was totally removed. There was moderate residual left hemiparesis that cleared completely by two weeks after the operation. The patient was discharged on anticonvulsant medication.

One can only speculate as to the reasons why this

benign lesion, which had been present in a "silent" state for months or years, suddenly became an acute clinical problem that required complete investigation and surgical treatment. The successful outcome in this case illustrates the need for maintaining sensitivity to the possible existence of surgically remediable lesions even in patients of advanced age with multiple systemic disease.

Case #3. An 80-year-old man was well until two months prior to the first neurologic examination. At that time he was assaulted, and rendered unconscious for several minutes. When seen one day after the injury, there had been no significant neurologic signs. Two months after the injury he telephoned his physician and stated "I've had a stroke." When seen by the neurologist at that time, a history was obtained that he had begun to experience bifrontal head pains three weeks previously, and had noticed some weakness of the right hand. Three days prior to the examination the right hand had become "numb," and he was unable to grasp objects. His speech became affected in a way that was described as "I want to tell my wife something but the words will not come out." His gait had also become slower. The neurologic examination at that time revealed a mild organic mental syndrome. No gross aphasic defects were noted. There was a right facial weakness, and weakness of the right extremities. There were no reflex changes. Sensory examination was normal except for extinction of touch stimuli to the right side under conditions of double simultaneous stimulation. Plain skull x-rays were normal. The EEG revealed left-sided focal dysfunction. Spinal fluid chemistries and pressure were normal. A brain scan suggested a lesion involving the convexity of the left cerebral hemisphere. An angiogram showed a large subdural hematoma over the right cerebral hemisphere. On the day following the angiogram, the patient developed a definite left hemiparesis. He was treated with steroids and improved rapidly (14). Repeat angiogram performed three weeks later demonstrated a decrease in the size of the subdural hematoma. Nuclear scans done repeatedly during the next seven years revealed complete resolution of the hematoma, and a CT scan done seven years after the incident showed no mass present (Fig. 3).

This case shows how separate bilateral lesions were demonstrated, to varying degrees, by both clinical phenomena and by different laboratory methods. It also shows how noninvasive methods can be used in following the course of patients during the recovery period.

Case #4. A 65-year-old man was admitted to the hospital because of a convulsion. Three hours there-

after, he was seen in neurologic consultation and it was noted that he had a severe organic mental syndrome and a mild but definite right hemiparesis.

He gradually improved and the neurologic workup revealed an abnormal electroencephalogram with a focus in the mid left cerebral hemisphere, a normal nuclear scan and a CT scan which showed a hemorrhagic lesion in the left posterior cerebral hemisphere surrounded by an area of edema (Fig. 4).

He gradually improved and was able to be discharged from the hospital. One month thereafter, a repeat CT scan revealed that the lesion was much smaller and seemed to be resolving (Fig. 5). Clinically, the patient was asymptomatic and had no findings. The impression at that time was of an encephalopathy caused by vascular disease with an intracerebral hematoma. His symptoms recurred, however, and four months later he was admitted to another hospital where he was found to have a glioblastoma in the same region.

This was a patient with an acute lesion that improved. The diagnosis seemed clear and he was discharged from the hospital without angiography and its potential morbidity. He did well and the repeat outpatient CT scan seemed to confirm the diagnosis. However, the diagnosis was incorrect. The histopathology following a decompressive craniotomy for neoplasm in the same region was glioblastoma multiforme. Retrospectively, it seems most likely that the initial clinical problem was due to hemorrhage into the neoplasm. In spite of the incorrect diagnosis, however, it must be emphasized that the patient was not harmed by the delay in treatment, considering the nature of the pathology and the clinical course.

Case #5. A 68-year-old man was admitted to the hospital for the sudden onset of "dizziness." By this he meant a sensation of imbalance which lasted about 20 minutes. Neurologic symptom review revealed that he had had headaches for one week prior to this and he had a "seizure" six months previously. At that time, he had been admitted to another hospital where the spinal fluid, EEG, and CT scan were all normal. Because of the concurrent presence of abnormal cardiac rhythms the final diagnosis was: 1) possible seizure disorder, 2) ventricular arrhythmia, 3) status postmyocardial infarction (which he had had three years previously).

The neurologic examination after the acute onset of imbalance revealed a normal mental status, a right facial asymmetry, a slight broad based gait but a negative Romberg test.

The impression was posterior fossa dysfunction,

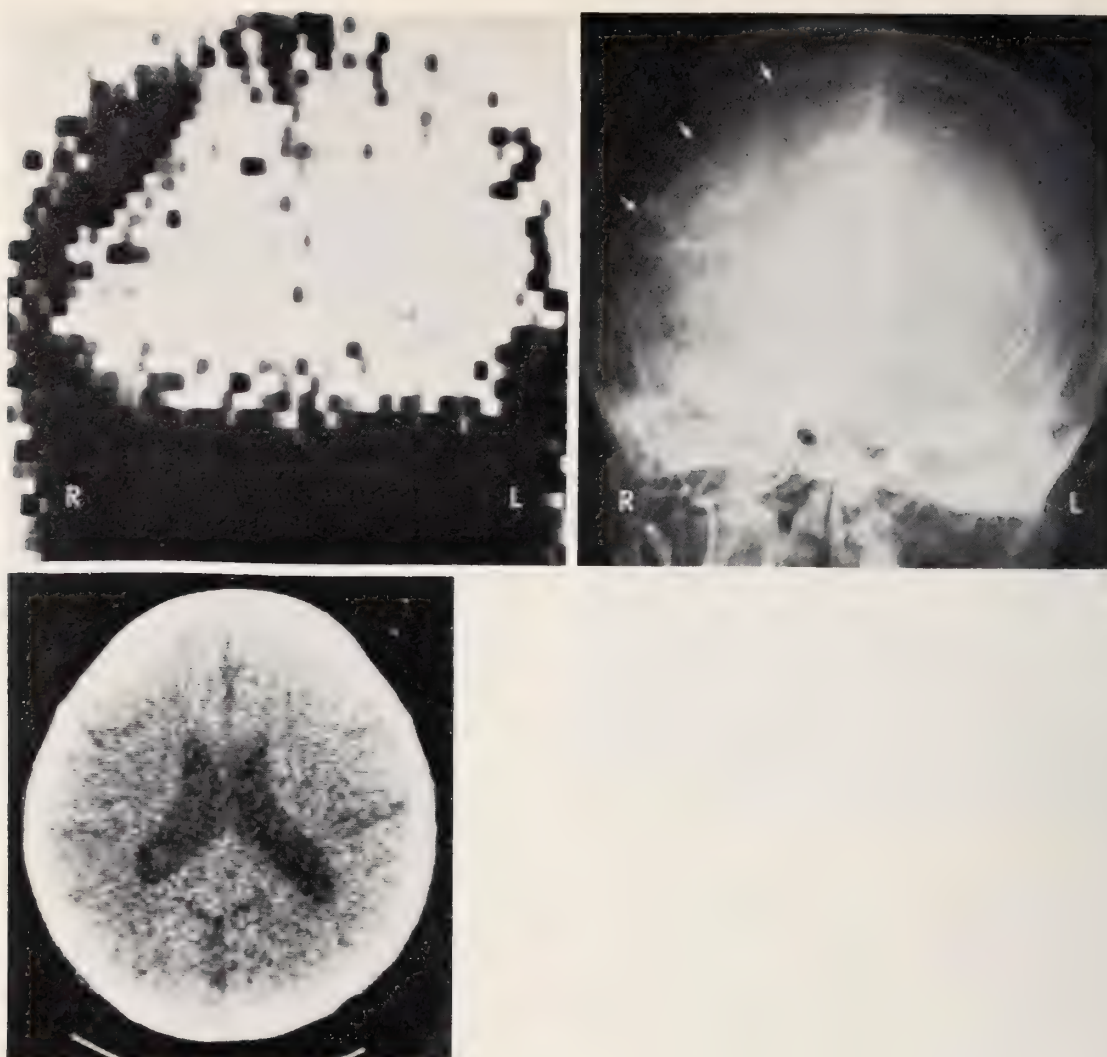


FIG. 3. a: Nuclear brain scan in anterior view showed at that time a crescent type of uptake consistent with subdural hematoma on the right. b: Cerebral angiography at that time revealed a large right sided subdural collection (arrows) with shift of anterior cerebral arteries. c: Precontrast CAT scan performed in June 1977 demonstrates complete resolution of the hematoma. (Reprinted with permission from Vol. 2, No. 4, p. 465, 1978.)

most likely involving the vestibular system. The etiology could not be determined by clinical examination but was thought to be vertebra-basilar insufficiency. In view of the complaint of headache, and the seizure history, studies were performed, and a CT scan revealed a lesion involving both frontal lobes at the region of the septum pellucidum (Fig. 6). Angiography demonstrated a glioblastoma multiforme.

This case demonstrates that a CT scan can be normal when done too early in the course of an illness. It is not clear whether the apparent vestibular system dysfunction was caused by the brain tumor or whether it was coincidental and a result of vertebrobasilar artery insufficiency. If the latter is

true, the CT scan picked up a relatively asymptomatic glioma.

The cases cited indicate circumstances and management methods in only a few of the infinite number of complex and often puzzling problems presented by patients in the geriatric age group. One thing they have in common, however, is that the patients were all relatively intact at the time of onset of illness. This circumstance justifies a fairly aggressive, although rational approach to the choice of diagnostic technique as described earlier in this paper. It must be recognized that this is not always the case. The premorbid level of functioning can be of determining significance in the physician's approach. A mechanical formula that matches tests to

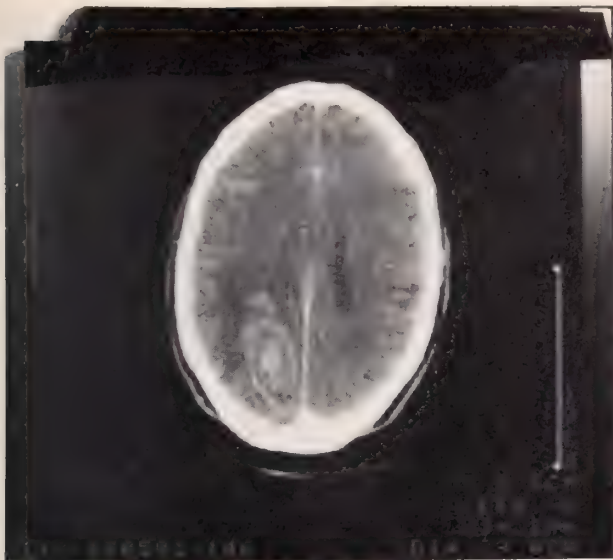


FIG. 4. Left posterior cerebral lesion.

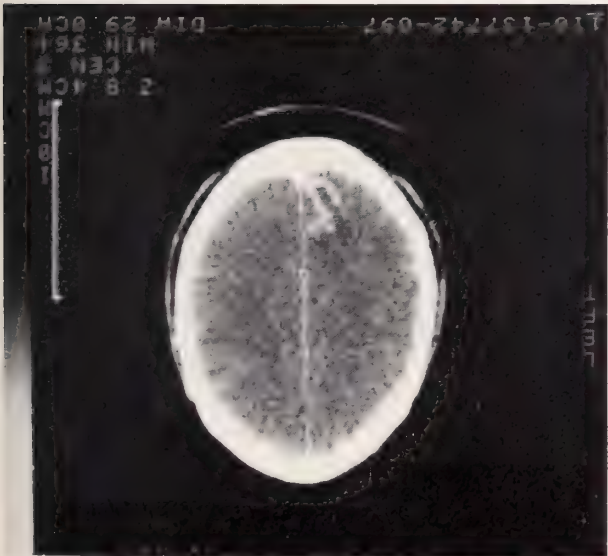


FIG. 5. Resolving left posterior cerebral lesion.

symptoms is unacceptable for many reasons, both philosophical and practical. A chronically demented institutionalized person with an acute hemiparesis cannot be considered equivalent, in terms of diagnostic strategy, to an elderly person who is fully active in the community and who has the same acute manifestations. Humanitarian as well as practical considerations may dictate simple supportive care for one and aggressive investigation for the other. The line between potentially helpful activity and needless meddling may be blurred at times, but the experienced physician must always try to distinguish one from the other.

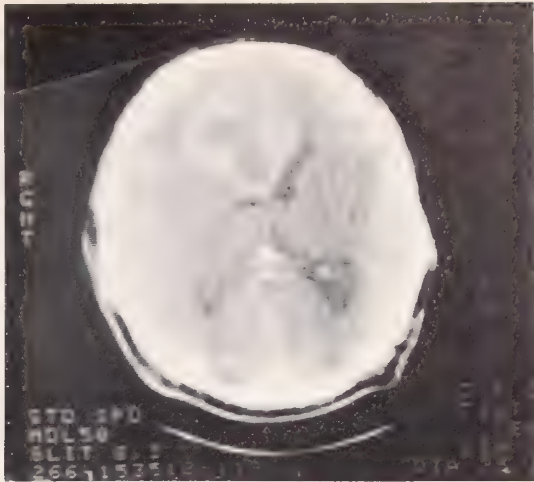


FIG. 6. Septum pellucidum lesion.

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Guillain-Barré Syndrome Following Trivalent Influenza Vaccine in an Elderly Patient

RICHARD BROOKS, M.D. AND ANDREA REZNIK, M.D.

Abstract

The Guillain-Barré Syndrome (GBS) has been described following many vaccinations, including most recently the swine flu vaccine. A case of GBS is described in close temporal relationship to administration of currently available trivalent flu vaccine in a geriatric patient. The implications of this phenomenon and a review of the literature are presented.

The Guillain-Barré syndrome (GBS) has been described following a variety of vaccinations including rabies (1, 2), measles (3), smallpox (4, 5), polio (6), and DPT (3). Interest in a possible association with influenza developed following mass immunization with monovalent swine flu vaccine in 1976. An increase incidence of this syndrome was noticed in recipients of the vaccine. We recently saw an elderly patient who developed GBS following administration of currently available trivalent influenza vaccine A Russia/ A Texas/ B Hong Kong.

Case Report

A 75-year-old woman was in good health until December 6, 1978, three weeks after receiving a trivalent flu vaccine on November 12, 1978, when she began to complain of weakness in both legs. She had a history of mild diabetes untreated, and hypertension treated with propranolol. There were no allergies.

Neurologic examination on admission showed severe weakness both proximally and distally in the lower extremities without atrophy, wasting, or fasciculations. The upper extremities were of normal

strength. Sensory examination was intact in the upper extremities, but showed decreased vibratory sensation in the lower extremities below the knees. Reflexes were absent in the lower extremities and plantar responses were flexor. The patient underwent a total myelogram to exclude the possibility of spinal cord compression which was normal. The spinal fluid protein was 115, glucose 78, with 9 red cells and 2 white cells; opening pressure was normal. Electromyography showed normal conduction velocities and findings suggestive of diffuse neuropathic dysfunction, runs of fibrillations and positive sharp waves. In the hospital, her condition worsened to the point of paraplegia with absent reflexes in the lower extremities. Her muscle power gradually began to improve, and she was finally discharged to a rehabilitation facility.

Discussion

The case described is typical of GBS. Progressive motor weakness and areflexia, as required by the NINCDS criteria for the diagnosis of GBS, were present (10). There were also many strong supportive features such as progression with relative symmetry, mild sensory symptoms, and elevated spinal fluid protein without pleocytosis.

Atypical, however, was the age of this patient. Although GBS has been described in patients older than 70 years of age (7), it is distinctly unusual.

Review of the literature reveals case reports of

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GBS following a variety of vaccinations. Many of the earlier case reports describe features considered atypical for GBS such as marked asymmetry of motor involvement (4) or spinal fluid pleocytosis (7). Other postvaccination complications clearly describe an encephalitis (8) or neuritis of a single nerve or nerve plexus (4).

Complications related to influenza vaccine were rarely reported until 1972 when several cases of encephalomyelitis (8) were described. In 1976, mass immunization with A/New Jersey, A/Victoria vaccine was initiated. The Center for Disease Control published data in December, 1976 (9) indicating an increased incidence of GBS seven times greater than that in the unvaccinated population. A moratorium on further vaccination was initiated at that time.

Influenza vaccine continues to be available, primarily for individuals believed to be at high risk for the serious complications of influenza. Unfortunately, it is these same individuals who are likely not to fare well with any neurologic complications. The risk of developing neurologic complications such as GBS cannot be assessed for the individual patient. Clearly, the rare association with oral polio or DPT vaccine might represent no more than a chance occurrence while the increased attack rate described following swine flu vaccine is more suggestive of an etiologic relationship. What is needed, and is currently in progress through the Center for Disease Control and the American Academy of Neurology, is a prospective study to determine what relationship, if any, there is between

vaccination and GBS. The currently available trivalent flu vaccine is predominately being administered to elderly or debilitated patients, as in the case described. This may expose a segment of the geriatric population to neurologic complications previously rare in this age range.

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Cervical Vaginal Screening in the Over 65 Female

STUART OSTER, M.D.

Abstract

This study was designed to evaluate the necessity of performing cervical vaginal smears on women over the age of sixty-five. Over a three-year period, there were 1130 smears performed on 420 asymptomatic women over sixty-five. This screening program revealed two women with smears positive for invasive squamous cell carcinoma of the cervix.

Introduction

At present, deaths from cervical cancer are predominantly among women who have never had a cervical smear taken. The screening test is designed to sort out apparently well persons who probably have a disease from those who do not.

There is some controversy over the age at which a woman should cease having a cervical smear taken. The Walton Report from Canada recommends regular cytologic testing be discontinued at the age of sixty (1). The British Society of Cytology advocates discontinuance of vaginal smears at the age of seventy (2).

In the United States, we have recommended cervical vaginal smears be repeated annually, on all adult women, until the end of their lives.

This study was designed to evaluate the necessity of performing cervical vaginal smears on women over sixty-five.

Materials and Methods

This study conducted from September 1976 to September 1979. The patients were all from two nursing homes in the New York area. The Jewish Home and Hospital for Aged which has two facilities, one in Manhattan and the other in the Bronx, with a combined total of 1440 beds. Approximately 75% of these beds are occupied by females. The

other nursing home, the Amsterdam House in Manhattan, has 415 beds with again 75% female occupancy.

The Jewish Home and Hospital female population is approximately 90% Jewish, with very few non-white patients. The Amsterdam House is over 90% non-Jewish, and approximately 8% non-white. The age distribution was the same for both homes. All patients included in this study were over sixty-five, with the median ages 70-80, and included patients over 100 years of age.

At the Amsterdam House, I did cervical vaginal smears on all the female patients at the home from September 1976 and when possible repeated the smears every year for the next three years. Any new admissions during this time had a pap smear. At the Jewish Home and Hospital, all patients referred to the gynecologic clinic, for whatever reason, had a pap smear taken. If the patient was referred to me again I did not repeat the cervical smear if she had the same complaint or a previous negative smear. Allowing for new admissions, transfers, and deaths, I did 1130 cervical vaginal smears at the Amsterdam House, 420 were initial tests and 710 were repeat smears. At the Jewish Home and Hospital 240 smears were taken on 220 patients and 20 were repeat tests.

Results

At the Amsterdam House, where all patients were screened by taking a cervical vaginal smear, two patients had smears positive for invasive squamous cell carcinoma of the cervix. The women aged 71 and 81 were both white, asymptomatic and had no

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previous knowledge of ever having a pap smear taken. Both positive smears were obtained on the initial screening. The 81-year-old patient refused further diagnostic workup and the 71-year-old lady's cervical biopsy revealed squamous cell carcinoma of the cervix. Both were Stage I.

At the Jewish Home and Hospital there were no positive smears. An 81-year-old patient came to the clinic with a procedentia. The prolapsed cervix looked suspicious and a biopsy revealed squamous cell carcinoma of the cervix Stage I-b.

In our study, three women over sixty-five years of age had squamous cell carcinoma of the cervix, two cases were asymptomatic and picked up in our screening program.

Discussion

Since the introduction of cytologic screening to the general female population four decades ago, there has been a changing incidence of cervical cancer with a decrease in the morbidity and mortality of invasive squamous cell carcinoma of the uterine cervix. The decrease in morbidity and mortality in most studies for squamous cell carcinoma has a close relation to cytologic screening activity. Cytologic screening of a population will significantly increase the proportion of patients with Stage I invasive carcinoma of the cervix, and will identify the bulk of those with preinvasive lesions (3).

Rescreening at yearly intervals will virtually eliminate invasive cervical cancer. Rescreening at longer intervals will significantly decrease the rate (4).

Invasive carcinoma of the cervix is primarily a disease of the second half of life, with the incidence of new disease (that is conversion from a negative to a positive smear) greatest at ages 25-29 and the maximum prevalence at ages 35-39. We define a positive cervical smear as one that has cells from severe dysplasia, carcinoma in situ or invasive cancer of the cervix (5).

In the much quoted British Columbia figures for age-specific incidence of conversion of negative to positive cervical smears, the ratio fell from .39-1000 persons rate at age 55-59, to .21 at 60-64, .11 at 65-69, .08 at 70-79, and .00 at 80- and older (2).

The Walton Report from Canada concludes that cervical neoplasia is rarely detected cytologically in women aged 60 and over. The report recommends that regular testing at this time be discontinued (1).

Spriggs and Housain from Great Britain in a paper prepared for The British Society of Clinical Cytologists recommends regular screening seems inappropriate after age 70 though a first smear should always be taken from an unscreened woman at whatever age (6).

E.R. Siegler, in 1967, at the annual meeting of The American Society of Cytologists reported his findings on 483,000 pap smears over a ten-year period. Thirty thousand were women over sixty-five. He reported 33 had invasive carcinoma of the cervix (7).

It appears from our study that there are women over 65 with undiagnosed cancer of the cervix. Therefore, an initial pap smear is indicated on all patients regardless of age. Both positive smears in this study were on previously unscreened patients, proving it is necessary to screen all women over 65. Those who test negatively will more than likely remain negative. Conversion is unusual past 65. Cervical smears in those females over 65 is more significant, because as this study indicates, positive smears are associated with advanced cervical disease at this age.

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Otolaryngologic Care of the Elderly

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The most common complaint of the elderly is impairment of hearing (1-4). The very essence of living requires facile communication with one's fellow beings. When hearing deteriorates, people become irritable and withdrawn. This condition may progress to isolation, suspicion and even paranoid reactions (3).

In more than half of the patients first seen, the hearing impairment is caused by an accumulation of cerumen and keratin from desquamated epithelium. Removal requires care to avoid injury to the skin of the canal and tympanic membrane. It is facilitated by softening the area for several days with glycerine and peroxide prior to irrigation with hypertonic saline at body temperature. Perforations of the tympanic membrane and previous mastoid surgery are contraindications to irrigation. In these instances, gentle removal with suction and a blunt ear curette is preferred. After cleansing the ear canal, the hearing is tested. Although emphasis has been placed on audiometric studies, it is also important to test for speech discrimination. Tuning fork tests are helpful to determine the relative impairment for conduction and sensorineural function. Surgical correction of conductive hearing defects in selected cases may be feasible. Surgery is restricted because of added risks, concomitant medical conditions, and limited life expectancy. The proper fitting of hearing aids and instruction in their use afford some relief to the patient. Training in lip reading is also essential. Presbycusis, or normal hearing loss attributable to age alone, is characterized primarily by degenerative changes in the cochlea. Other lesions occur in the middle ear, auditory nerve, cochlear nucleus and at various levels of the central nervous system (4).

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Dizziness is the second most frequent complaint. Normal equilibrium is a state of harmonious symmetry between the right and left vestibular systems as they function to maintain the proper relationship between the person and the environment. Vertigo denotes a specific illusion of motion related to turning or falling. Dysequilibrium is used to describe symptoms of spatial disorientations. Central vestibular disorders are most commonly due to vascular lesions (4). Neurologic examinations are indicated in these instances. Questioning the patient will usually elicit a history of fear and anxiety. Figuratively, their "world is turning upside down." It is advisable to avoid excesses of food, salt, coffee or tea, and smoking tobacco. Useful drugs include Dimenhydrinate (Dramamine) and Meclizine (Antivert). Medication for other conditions should be carefully reviewed and unessential drugs eliminated.

Tinnitus is an annoying symptom, but in most instances can be managed by teaching the patient to ignore it. There is no effective medical treatment. Masking devices may be used to allay the discomfort.

Pain in the ears without otologic pathology is frequently caused by malocclusion caused by changes in the teeth, gingivae and supporting bony structures, as well as outworn ill-fitting dentures, resulting in temporomandibular dysfunction (2).

Dryness of the nasal mucous membranes gives rise to a sensation of stuffiness. Proper humidification of the environment affords some relief. Intranasal medication is not recommended.

Nose bleeds in the elderly are troublesome. They are frequently precipitated by worry or conflict. Control of epistaxis requires patience, gentle and limited chemical cauterization, and anterior packing. Postnasal packs have been reported to cause fatalities due to anoxia.

In the throat, the diminished function of the salivary glands and the muscles of deglutition result in difficulties in swallowing. These are ameliorated by teaching the patients to take small bites, to chew

food thoroughly and to swallow their salivary secretions. A common error is unnecessary use of anti-septic mouth washes and gargles. Difficulty in speaking arises from faulty innervation of the muscles of phonation (2). This can be alleviated by vocal exercises.

Naturally, elderly patients are subject to other causes of disease. The incidence of malignant tumors in the patients over 75 years of age is lower than the general population.

The elderly are prone to frequent falls. In most instances, these result in fractures of the extremities. Nasal fractures are relatively infrequent. When the nasal fracture results in impaired respiration, or significant deformity, it may be corrected by closed reduction under local anesthesia.

In the treatment of the elderly a paradoxical situation has developed in recent years. Since the advent of Medicare, the zealous efforts of physicians to prolong existence, and the demands of the patients to "do something, doctor," have resulted in widespread over-medication. In one study, it was reported that 87% of the patients were on regular drug therapy, a figure three to four times than in the general population. One patient was receiving 13 medications (5). The multiplicity of the drugs results in unfavorable drug interactions. Elderly patients benefit more from the physicians stopping drugs rather than starting them (6). The decreased ability of the liver to metabolize the drugs and the decreased ability of the kidney to excrete them require that the dosage of drugs be reduced (7).

A contributing factor to hearing impairment and equilibratory dysfunction in the elderly is medication (1, 3, 8). While the ototoxic effects of quinine and streptomycin have been widely recognized, the damage to the auditory and vestibular organs by a variety of other drugs has not been generally appreciated. The drugs used in the elderly to treat severe infections, arthritis, hypertension, and other medical problems, may have deleterious effects on the inner ear (8, 9).

Foremost of the ototoxic drugs are the aminoglycoside antibiotics. These include amikacin, dihydrostreptomycin, gentamicin, kanamycin, neomycin, streptomycin, tobramycin, and vancomycin. The toxicity is related to the increase in concentration and duration of time during which the antibiotic is retained in the inner ear fluid. The vestibular damage caused by gentamicin may be reversible in half of the cases if diagnosed early (9). In vestibular toxicity from streptomycin, the damage is reported to be irreversible. Ninety percent of patients treated

with daily doses of 2 gm for 30 days showed signs of toxicity (10). Neomycin ototoxicity has been reported to occur in patients when it was used solely as a bladder irrigation (11).

Among the analgesics, salicylates account for a significant number of cases of ototoxicity (12). Impaired hearing and tinnitus have also been reported from the use of ibuprofen (Motrin) and indomethacin (Indocin) (9).

Diuretics that are given over long periods of time to most elderly patients may be ototoxic. This is believed to be from alteration of electrolyte composition in the inner ear fluids. Ethacrynic acid (Edecrin) (13) and furosemide (Lasix) (14) are examples.

The antineoplastic agents linked to ototoxicity include bleomycin, dactinomycin, diamminodichloroplatinum and mechlorethamine (Nitrogen mustard) (8). Other drugs reported to be ototoxic are chloroquin (Aralen), lidocaine (topically to the ear), colistin (Colymycin), polymyxin B (Aerosporin), propranolol (Inderal), and quinidine (9). Sulfamethoxazole (Gantanol) and Trimethoprim (Bactrim, Septra) are reported to cause mild reversible impairment in hypertensives (11).

To minimize the chances of ototoxicity, the following guidelines are suggested:

- (a) The lowest possible effective dose should be used.
- (b) Prolonged administration should be avoided.
- (c) Patients should be monitored for signs of ototoxicity.
- (d) They should also be monitored for renal and hepatic function, and the dose of the drugs adjusted accordingly.
- (e) Patients should be well hydrated to avoid ototoxicity from concentration in the inner ear fluids.
- (f) Patients with existing hearing loss should be monitored closely, and if possible, potentially ototoxic drugs should be avoided (8, 9).

Summary

The causes of complaints in the aged are primarily degenerative changes throughout the body. The changes are aggravated by excessive medication. Drugs should be given only when necessary, under careful supervision, in reduced dosage, and for limited periods. Surgery should be severely restricted. The patient's wishes should be scrupulously respected.

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Progress in Ophthalmology for the Elderly since the 1971 White House Congress on Aging

ABRAHAM L. KORNZWEIG, M.D., F.A.C.S.

On November 28, 1971, a White House Conference on Aging was held in Washington, D.C. (1). The theme of the conference was directed toward a national policy on aging. Special sessions were arranged for special problems. Among these was a special session on aging and blindness. A copy of the program of that session is shown in Figure 1, to indicate the extent of the discussion. Toward the end of the session, final recommendations were voted on and adopted. The second recommendation for improvement and maintenance of the health of the eyes, and prevention of eye diseases in the elderly is the following:

"It is recommended that the National Eye Institute and other interested organizations on a national and local level combine their efforts in an urgent overall program to prevent and alleviate diabetic retinopathy, establish a center for the study of diseases of the macula and increase research efforts in the fields of cataract, glaucoma and vascular diseases of the eye; establish screening efforts especially at hospitals, medical centers, homes for the aged, nursing homes and extended care facilities to find aged patients who have blinding eye diseases which can be helped by medical and surgical means and low vision aids. Such efforts should be made by interested philanthropic organizations and implemented if necessary by legislative action. It is further recommended that the

National Eye Institute be required to develop better statistics on incidence, prevalence and etiology of blinding eye conditions."

In addition to the special session on aging and blindness, a general policy statement from the health committee of the White House Conference on Aging was also issued;

"Among the complex of health needs of the aged, priority should be given for the prevention and treatment of blindness."

Other recommendations had to do with social and economic problems, vocational rehabilitation, transportation and home care.

This paper will present the most important advances in ophthalmology for the aged since the last White House conference in 1971. Special attention will be given to the recommendation of the congress to see if they were fulfilled.

A Diabetic Retinopathy Research (DRS) Study was initiated by the National Eye Institute with a federal grant in June 1973 (2). Sixteen eye centers were picked, and a carefully planned and controlled study of the effects of laser beam photocoagulation to the periphery of the retina on the progression of the disease. The photocoagulation produces an ablation of this part of the retina. The objective was to see if this treatment was useful in preserving vision for patients with proliferative diabetic retinopathy. This was a clinical experiment on a national scale. Previous experience on the use of the laser beam to treat hemorrhagic areas of retina have proved helpful in eliminating the hemorrhage. This led to the theory that broader areas of the retina treated by laser photocoagulation might prevent the occurrence of hemorrhages in the central or macular area, which is so important for clear and sharpest vision. Several individual ophthalmologists in the United States and abroad reported good

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1971 WHITE HOUSE CONFERENCE ON AGING

PROGRAM

AGING AND BLINDNESS

GARSON MEYER, *Presiding*

8:00 A.M. OPENING REMARKS

- DR. PETER J. SALMON, Trustee, American Foundation for the Blind, Inc., New York, N.Y.
- DR. ROBERT MORRIS—GARSON MEYER, Co-Chairmen of the National Task Force on Geriatric Blindness, American Foundation of the Blind
- DR. WILFRED D. DAVID, Executive Director, National Society for the Prevention of Blindness, Inc.
- DR. ROBERT A. RESNIK, Chief, Office of Program Planning, National Eye Institute, Bethesda, Maryland

8:30 A.M. SPEAKER

- SENATOR JENNINGS RANDOLPH, West Virginia, Special Committee on Aging, U.S. Senate, *Special Needs of the Aging Blind Population*

9:00 A.M. PANEL

- Policy and Platform Statement in Behalf of the Elderly Blind Population in the U.S.*
- Income Maintenance
DR. JUANITA M. KREPS, Professor of Economics, and Dean, The Women's College, Duke University, Durham, North Carolina
- Medicine and Health
DR. A. L. KORNZWEIG, Chairman, Liaison Committee American Foundation of Blind, American Geriatrics Society, New York, N.Y.
- Rehabilitation
DR. DOUGLAS C. MacFARLAND, Director, Of-

fice for the Blind and Visually Handicapped, Social and Rehabilitation Service, U. S. Department of Health, Education, and Welfare, Washington, D. C.

—Transportation

WILLIAM C. FITCH, Executive Director, National Council on the Aging, Washington, D. C.

—Home Help Service

DR. ROBERT MORRIS, Director, Max Levinson Gerontological Policy Institute, and Professor of Social Planning, the Florence Heller Graduate School for Advanced Studies in Social Welfare, Brandeis University, Waltham, Massachusetts.

10:40 A.M.—Discussion of Policy Recommendations

An invited group of elderly blind persons and special personnel will serve as reactors during this period.

—Invited Reactors

WILLIAM EDWARDS, Washington, D.C.; MILTON KLEIN, Columbus, Ohio; HAZEL LEMKE, Rochester, New York; APARICIO G. RANGHLE, Washington, D.C.; DURKWARD K. McDANIEL, National Representative, American Council of the Blind, Washington, D.C.; and JOHN F. NAGLE, Chief, Washington Office, National Federation of the Blind.

11:00 A.M.—Adoption of Policy and Platform Statements

By floor vote for final recommendations in the White House Conference Report.

12:00 Noon—Adjournment

FIG. 1. Program, White House Conference on Aging, November 28, 1971.

results with this method in retardation of diabetic retinopathy and preservation of central vision. The National Eye Institute and its council of advisers then decided to give this method of treatment a national trial. Other objectives were to determine at which stage the treatment was most helpful, whether the treatment was harmful at any time, which form of photocoagulation was better, the argon laser beam or the zenon arc, and finally to study the natural progression of the disease without treatment. About 100 patients who had diabetic retinopathy were to be seen at these centers. One eye only was treated, the other eye was used as a control. Informed consent was obtained and the patient was to be under observation for five years. The preliminary results of the study were released in October 1977 (2).

As a result of this study, it was found that photocoagulation with the laser beam of the peripheral retina, when compared to indefinite deferral of treatment was of benefit in all stages of retinopathy included in the study, on the basis of:

1. reduction in the rate of visual loss.
2. slowing of progression of the retinopathy.

Some harmful effects were observed.

1. Reduction of peripheral field more marked in patients treated with the zenon arc laser beam
2. Loss of 1–4 lines of visual acuity in 10% of patients treated with the argon laser beam and about 20% of patients treated with the zenon arc laser.

When risk of severe visual loss without treatment is great, chance of benefit from treatment outweighs the risk of harmful effects.

For eyes in earlier stages, treatment or continued observation is equally acceptable at the discretion of the ophthalmologist and patient.

When risk of severe visual loss without treatment is small, risks of treatment deserve more serious consideration.

Continued follow up of DRS patients is essential,

1. to determine which of the two laser beams is better.
2. to evaluate long term results of treatment.

Also in a more recent report, this study has found the four risk factors which, if they are present in any case of diabetic retinopathy, will lead to severe visual loss (3). They are: 1) the presence of vitreous or preretinal hemorrhage, 2) the presence of new vessels, 3) location of new vessels on or near the optic disc, 4) severity of new vessel formation. The greater the number of risk factors in an eye, the greater is the possibility of severe visual loss in a two-year period.

This is a very valuable and practical clinical study of the subject of diabetic retinopathy, from which much has been learned, and a means of preventing and ameliorating the severity of the disease has been found. It should be continued for another five years to gather more information. There are side effects to this treatment in loss of vision and pe-

ripheral fields in some cases that are undesirable. Ablation of part of the peripheral retina is an heroic treatment, but better than complete visual loss.

A similar program on the effects of anticoagulants and of acetyl salicylic acid, to determine what these drugs may accomplish in the prevention or amelioration of diabetic retinopathy should also be considered. In addition, further research of basic chemical and pathologic studies are indicated, as was recommended by the White House conference, to learn more of the underlying metabolic factors in diabetes.

A serious complication of diabetic retinopathy is the recurrent vitreous hemorrhage that will eventually result in complete visual loss. These hemorrhages come from the proliferating blood vessels on the surface of the retina which are fragile and bleed easily. Efforts to treat this complication were initiated especially by Machamer in Florida. He devised an instrument to remove the hemorrhage by aspiration and cut the vitreous strands that were attached to the retina. The vitreous was then replaced by clear, sterile normal saline. Results at first were very limited, but improvement came with more experience and better instrumentation (Fig. 2) (4). At the present time, a completely new surgical procedure has been created called vitrectomy (5).

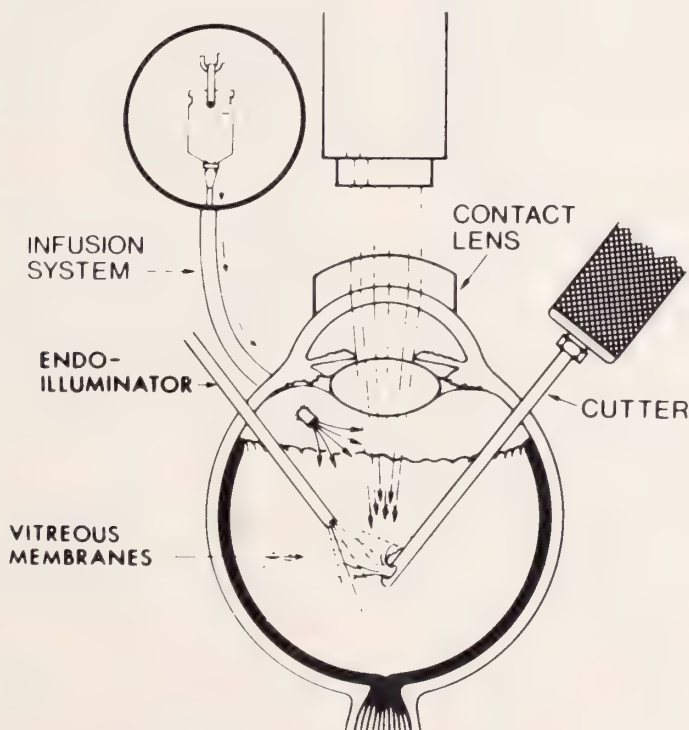


FIG. 2. Diagrammatic representation of instruments involved in the vitrectomy operation.

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Its use has been expanded to include hemorrhages in the vitreous from other sources such as trauma and complete occlusion of the central retinal vein. Intraocular foreign bodies and parasites can now be removed by direct observation and the illumination is provided by a fiber optic probe. These instruments penetrate the eyeball through a region of least vascularity, the pars plana of the ciliary body. This area is located in the anterior segment of the eye, 3-7 mm from the limbus of the cornea, and it completely encircles the globe. Vitrectomy is another way of maintaining vision in the affected aged patient—thus, carrying out the recommendation of the White House Conference.

Perhaps the most interesting and exciting procedure in this decade is the reintroduction of the intraocular lens implant after cataract extraction. About 30 years ago, Ridley in England tried it, using a plastic lens to replace the cataract that was removed. At that time the complications were too numerous. Too many eyes were lost and the operation was abandoned. But more recently, Binkhorst in the Netherlands, using a more sophisticated technique and a lighter, carefully sterilized methyl methacrylate lens was much more successful. The operation caught on. Other ophthalmologists in England, Europe and America devised different types of intraocular lens implants and attempted to improve on previous methods (6-9). The success rate has now reached 80%-85%. Some eyes are not amenable to this type of surgery, specifically diabetics, high myopias, glaucoma cases, infectious eyes with uveitis or iritis. It appears to be most satisfactory for the aged patient, especially the person who has difficulty adjusting to the usual cataract glasses, and who is unable to wear a contact lens. The plastic lens remains permanently inside the eye as close to the original position as possible. Some lenses are designed to remain in front of the iris, others in the plane of the iris, and finally behind the iris, which is the original position of the normal lens. Complications do occur and some eyes have been lost because of infection. The Food and Drug Administration has stepped in to monitor the procedure. It now requires the surgeon to be certified as an investigator and to report periodically every three months on the results of surgery. At the end of a year, the results of all cataract extractions have to be reported. Manufacturers of the plastic implants have to ensure the sterility of the lens and of the solutions in which they are kept, to avoid post-operative infections. Still, in spite of the complications, successful patients are enthusiastic, because they can see clearly almost immediately after the

operation, and may not need glasses except to read. They do not have to use contact lenses, which are difficult to insert and remove and have to be kept constantly clean and sterilized, a difficult job for many older patients. Informed consent has to be signed by the patient or next of kin, after being told of the nature of the procedure and the more usual complications.

The latest report of the Food and Drug Administration (10) stated that there is a limited but alarming incidence of complications from intraocular lens implants based on the first nine months of study. This information on approximately 70,000 lens implantations and 1,500 control cataract operations is shown in Table I.

Successful patients tell their friends and relatives and, in spite of complications, some patients are beginning to ask for the lens implant operation. Eventually, the best patient and the best procedure for the lens implant surgery will be found. The remainder will use the more customary methods of cataract extraction.

One of the recommendations of the special session on aging and blindness was the establishment of a center for the study of diseases of the macula at the National Eye Institute. A center, as such, has not been started as yet, but considerable research has been done there and at other eye centers funded by grants from the National Eye Institute.

Degeneration of the macula in the eyes of aged patients affects about 24% of all patients over the age of 65. It increases with age to 38% in the group over 80-years old (11). The macula is the area of sharpest and clearest vision. It enables the person to see clearly in the distance (so called 20/20 vision) and to identify friends and relatives. It is used to read find print and sew fine stitches. It works best in daylight, and is called daylight vision in contrast to peripheral vision which works best in dim light and is called night vision. A small area in the retina, it is about $\frac{3}{8}$ of an inch in diameter situated at the posterior pole of the eye on the temporal side of the optic disc. As the macula degenerates, vision is

TABLE I
Complications Following Intraocular Lens Implant Surgery

Complications	Percentages of Adverse Reactions	
	Control	Lens Implants
Hypopyon	0.21	0.31
Intraocular infection	0.06	0.13
Acute corneal decompensation	0.00	0.17
Surgical reintervention	0.26	1.40
Total	0.53	2.01

reduced to 20/40, 20/70 and finally 20/200 or less. At that stage, the patient is severely handicapped visually, and is called industrially or legally blind. Such a patient is not totally blind. His peripheral or side vision is intact, and these patients can get around, feed and clothe themselves and even travel alone. But they cannot read a newspaper or a telephone book, sew on black material or identify a person at a distance. It has been compared to a person looking at a mirror on which the silver on the back of the mirror has been scraped off in the center, so that there is no reflection from this area, and there is a central blind spot.

Like cataract, macular degeneration is believed to be the result of the aging process involving mainly the cone cells of the retina, the adjacent pigment epithelial layer, Bruch's membrane, which separates the retina from the choroid, and the chorioidal capillary layer which supplies the circulation to the macular area.

Macular degeneration in the newborn and the young is hereditary and there are many family pedigrees to prove this fact. Some ophthalmologists believe that this is also true of macular degeneration in the aged. It is much more difficult to prove this assertion. There are only a few incidences in the literature that cite the occurrence in members of a family, or in twins (12). Besides in the aged, there are environmental and general medical conditions that may contribute to degeneration of the macula. Excessive light as from the corona in an eclipse of the sun will cause a permanent macular scar in the eye of an individual who will look at the eclipse without adequate protective lenses. Also, the bright light from a welder's arc in industry will do the same to the macula if the eyes are not protected. Experimentally, mice exposed to ordinary light over prolonged periods have shown degenerative changes in the photoreceptor cells of the retina. General diseases such as diabetes, hypertension and kidney diseases affect the retina and the macular area. Certain parasitic diseases such as toxoplasmosis, histoplasmosis and amoebiasis have been known to involve the macula (13).

It has been found experimentally that the tips of the retinal photoreceptor outer segments (rods and cones) are continually being removed by filaments of the retinal pigment epithelium and phagocitized by the retinal pigment cell. These tips are then replaced by fresh tissue from the rod and cone cell body (14, 15). This metabolic exchange has led to much speculation and study as a possible cause of macular degeneration in the aged. The cells of the pigment epithelial layer have a great burden during

life of disposing of the cast-off photoreceptor membranes, that function well in the early years but could become more difficult in the aged. This could lead to drusen formation in the retina and areas of residual tissue beneath the pigment layer, accumulation of degenerate material in Bruch's membrane, and interference with the metabolic exchange between the retina and the choriocapillary circulation which is the main source of supply to the macula and foveal region of the retina (16).

Further research in this area is in progress. An animal model has been found in the rhesus monkey that has a macular resembling the human and that could be used for experimental purposes. Also, a research project involving human donors in a home for aged who have macular degeneration and have donated their eyes after death to the eye research laboratory is in progress. This would make available eyes that are as close as possible to the living tissue that can be studied by the electron microscope and tissue culture technique.

There is no reliable treatment at present for macular degeneration in the aged. Excessive vitamins, low fat diets, vasodilators, even small doses of corticosteroids over long periods have been tried with minimal success. The greatest help has come from low vision aids and clinics. These provide magnifying glasses to enlarge the image and other accessories to enable such patients to function better. Enlarged print newspapers and books are used. Closed circuit television units are available—though expensive—to enable the patient who can manipulate the controls to read books, letters and other material.

Institutions for the aged, and senior citizen centers that can afford them could purchase these instruments for use by many individuals. Radio reading services have been established in many states. These are special radios on separate channels that are used by the visually handicapped or the physically handicapped or home bound person who is unable to read or get a newspaper. The special radio will provide daily news coverage, stories, trade and financial news and other interesting material. The radio is not expensive and in many places, is provided without charge to deserving handicapped persons. Some of these programs are on the air for 20 hours, others at regular intervals. More information on these services can be provided by the Association of Radio Reading Services at 15 West 16 Street, New York, N.Y. 10011. Another organization involved in this service in the New York metropolitan area is called "In Touch," situated at 322 West 48 Street, New York, N.Y. 10036.

The recommendations of the White House conference include an increase in the numbers and greater utilization of low vision clinics. The American Foundation for the Blind has undertaken this project and now has a list of all low vision clinics in the country that is available to anyone who asks for it. The Foundation is located at 15 West 16 Street in New York, N.Y. 10011.

The other recommendations from the White House Conference on Aging include screening efforts to find the aged patient that is visually handicapped who may be residing in homes for aged, nursing homes and extended care facilities, and who could be helped by medical or surgical means and low vision aids. This effort has been essentially in the hands of the National Society for the Prevention of Blindness and its many branches across the country. Individual screening efforts by local groups or persons have been difficult to evaluate. All such efforts are valuable and should be continued. Incidentally, at Mount Sinai School of Medicine, a senior student inaugurated such a screening program at a senior citizens center close to the Elmhurst General Hospital. She found a number of patients who required medical and surgical eye care who were referred to the Elmhurst Eye Department. She was good enough to ascribe her interest in this subject to reading an article which I had written on the prevention of blindness for the aged for the White House Congress in 1971.

Basic research efforts should be increased in the fields of cataract, glaucoma and vascular diseases of eyes. This was also included in the recommendations of the special session on aging and blindness. Such research is in progress at the National Eye Institute, or parcelled out to individuals at different hospitals and eye centers. Definitive results are still not available. But the research program for the next few years was published by the N.E.I. in 1977 and is available (18). The National Eye Institute is located among the National Institutes of Health in Bethesda, Maryland.

Glaucoma is another eye disease that increases in frequency in the aged population (11). The percentage has been listed as 5%. If the number of cases of ocular hypertension is included it may go up to 9% or 10%. Ocular hypertension is found in many aged persons due to anatomic tissue changes associated with the aging process. The intraocular pressure increases above the normal, but the eye does not show evidence of optic nerve atrophy or loss of peripheral field in many cases. They have to be observed at regular intervals because some of these cases become true glaucoma patients with

loss of peripheral field and visual acuity. The main clinical problem in glaucoma is still to find the patient who has the disease but is unaware of it. For this purpose, as mentioned before, numerous screening efforts are performed on aged groups in senior citizen's centers and homes for aged. The National Society for Prevention of Blindness has been particularly involved in this field.

Both medical and surgical treatment is available to control the progression of this condition. Since 1971, newer surgical procedures have been devised to reduce the intraocular pressure when medical treatment alone is inadequate. They are called trabeculotomy and trabeculectomy that permit the aqueous fluid to drain off more readily into Schlemm's canal which is the usual outflow channel and, thus, reduce the intraocular pressure. These procedures are replacing the previous filtering operations that provided additional outflow channels into the subconjunctival space. Also, the argon laser beam is being used experimentally to create an opening in the iris to improve the flow of aqueous from the posterior to the anterior chamber of the eye (17). This opening converts a narrow angle into a normal wide angle anterior chamber and prevents the occurrence of an attack of acute narrow angle congestive glaucoma. It replaces the peripheral iridotomy procedure because it can be done in a clinic on an ambulatory basis, and is noninvasive surgically. Finally, a new drug has been introduced by the Food and Drug Administration for the local treatment of chronic open angle glaucoma (19). It is called timolol, and is a beta-adrenergic blocker. Its effect appears to be on the ciliary body by reducing the production of the aqueous fluid. Timolol is especially helpful for the aged patient, because it does not contract the pupil, or cause spasm of accommodation. Many aged patients have some cataract formation in addition to the glaucoma. A contracted pupil—which happens with the usual drug pilocarpine—will cause the cataract to cut down on visual acuity. A normal pupil may allow light to bypass the lens opacity. One drop of 0.5% or even 0.25% twice a day will control the intraocular pressure. All this has taken place since 1971.

In addition to the above, many new instruments have been devised to assist in diagnosis and treatment. Chief among these is the operating binocular microscope that has improved surgical procedures on the cornea, for cataract extraction and glaucoma operations. Newer and finer sutures, both synthetic and silk, 8-0 to 10-0, are used to close incisions more firmly, permit early ambulation, and prevent post-

operative infection and hemorrhage. More delicate instruments can now be used. The value of all these changes especially the binocular operating microscope cannot be overestimated.

Newer diagnostic techniques have been introduced. The A&B ultrasonic scanners have helped in diagnosing intraocular new growths, hemorrhages, retinal detachments and foreign bodies. The B scan is particularly helpful when the media of the eye is opaque, due to corneal opacities cataract formation or vitreous hemorrhages. The computerized axial tomography (CAT) scan has improved diagnosis of orbital and periorbital new growths and fractures in many problem cases.

New automatic refractors are available. Some are completely automatic and others are partly subjective. In the automatic refractor the patient looks at a fixed point and the result is obtained in a few seconds. In the subjective system, the patient participates in the process. In either case it is advisable to check the results at the trial case.

Fluorescein angiography, first introduced by Alvis and Novotny in 1963, is now a common procedure used universally in all cases involving the retina and choroid. It involves injection of 5% or 10% sterile fluorescein solution into the median cubital vein at the elbow. The patient is seated in front of a slit lamp that has a camera attached. The pupil of the affected eye is widely dilated. In about eight seconds, the dye is noted in the retinal artery, and photographs are taken in rapid succession for a minute and then at longer intervals, as the dye goes through the retinal circulation 10 or 15 minutes later. Much as been learned by this technique of disturbances in the retinal circulation in diabetic retinopathy, ocular tumors, macular disease and pigmentary atrophies and detachments. A modification of this test has recently been introduced in which the dye is given by mouth to avoid the occasional reaction of nausea or hives in some patients. The eye is examined 30–60 minutes later by the ophthalmoscope and photographed. This method is used in patients who have had an intraocular lens implant to see if a cystoid macular edema has occurred.

The laser beam has been mentioned before in the treatment of diabetic retinopathy and to perform an iridotomy. Its use has been expanded to seal off isolated retinal hemorrhages and tears, that may cause retinal detachments. A new project has been started by the National Eye Institute using the argon laser to seal off neovascular formations and hemorrhages in patients with senile macular degeneration. Thirteen eye centers have been assigned

this project, the results of which should be known by the next White House Conference on Aging in November 1981.

Noteworthy in the past decade, has been the improvements in contact lenses. Extended wear contact lenses are now available, mainly the soft contact lens. Patients have worn these lenses for three to six months without removal from the eye. This has been helpful for the aged person who finds it difficult to remove and replace the lens daily. These patients have to be observed frequently in the initial trial period to make sure that there is no corneal erosion or infection. A so-called bandage contact lens has been used to treat patients with painful corneal conditions such as bullous keratopathy, corneal ulcers, Fuch's corneal dystrophy, ocular pemphigus and corneal injuries. Much help in relieving pain has been reported by Refogo (20).

The decade 1971–1980 has been a period of considerable growth and development in ophthalmology and particularly for the aged. Fundamental causes of cataract, glaucoma and macular degeneration have not yet been found, but we look forward to the next decade with much interest and great expectations.

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Prostatic Problems in the Aged

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Urologic diseases constitute a significant percentage of the medical problems of the aged.

Urologic cancer accounts for approximately 25% of all new cases of cancer occurring among men, and accounts for approximately 15% of male cancer deaths. In females, cancer of the urinary tract accounts for 4% of all new cases and for 3% of cancer deaths.

Obviously, a detailed discussion of all the urologic problems affecting the aged is beyond the scope of this paper; it will, therefore, be limited to problems associated with benign and malignant diseases of the prostate.

Benign hyperplasia of the prostate is present in over 50% of males above the age of 50, and becomes clinically significant in increasing numbers with advancing age. Although most cases have a fairly classical clinical picture of nocturia, frequency, hesitancy, and diminution in the caliber of the urinary stream, a small, but significant percentage of patients, suffer from "silent prostatism" in which the patient is either unaware of his symptoms or ignores them because he feels that they merely represent part of the normal process of aging (1). These patients usually seek help because of overflow incontinence, uremic symptoms, or distended bladder symptoms that present as a pelvic mass.

The most common cause of hematuria in the prostatic aged male is benign hyperplasia of the prostate. Complete urologic evaluation is, of course, mandatory to rule out more serious causes of bleeding.

The intravenous urogram is of value in detecting abnormalities of the upper urinary tract, diverticuli, or tumors of the bladder, calculi and, to some extent, the size of the prostate. In addition, the amount of residual urine can be gauged quite accurately by the postvoiding film.

Cystoscopy is mandatory in cases with hematu-

ria, and usually can be performed just prior to surgery. In the absence of hematuria and with a normal intravenous urogram, preliminary cystoscopy is not always necessary.

The size of the prostate bears no relation to the degree or severity of the obstruction present, and is of significance only to the surgeon in making his decision as to which type of prostatectomy will be performed. The importance of the rectal examination (other than in determining the presence of rectal pathology) is to determine if the prostate is carcinomatous.

There is, as yet, no satisfactory medical treatment for benign prostatic hypertrophy. Sporadic reports of treatment with various combinations of estrogens and androgens have never proven to be effective. Most recently, some success has been reported from Israel in treating a small number of cases with Phenoxybenzamine (an alpha adrenergic blocker) but this has yet to be confirmed by other investigators.

Although the course is usually progressive, it is not always inexorable, and frequently may remain static for many years. Thus, while the need for a prostatectomy may be obvious in many instances (severe symptoms, large residual, upper tract dilatation, azotemia, calculi, recurrent infection, diverticula, etc.), there is a large group of cases in which symptoms are not too severe, and in which the disease may never progress to the point that a prostatectomy would be necessary. These patients may be followed for many years before surgery is required.

Often, a patient may request a prostatectomy, not because his prostatism is particularly severe, but because he feels that this will also eliminate the possibility of developing carcinoma of the prostate. It is important to inform the patient that the true prostate (in which carcinoma may arise) is left behind in all types of subtotal prostatectomy, and that total prostatectomy as a prophylactic measure in preventing carcinoma of the prostate has not been proven justifiable.

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There are four surgical approaches for removal of the prostate—transurethral, suprapubic, retropubic and perineal. Each has its own indications although they often may overlap, but all should yield the same result, that is, complete removal of the obstructing adenomatous tissue and a continent patient who empties his bladder with a full and forceful stream.

Cancer of the prostate is the most common malignancy in blacks and the second most common cancer in whites, and is found in 40%–60% of autopsies in men over 70 years of age (2). The lowest rates are found in Orientals in whom the disease is quite rare. Only 5%–10% of all cases of prostatic carcinoma are diagnosed early enough to justify radical prostatectomy in an effort to obtain a surgical cure.

Careful digital examination still remains the most reliable means of early detection of cancer of the prostate. An elevated acid phosphatase is indicative of disease that has spread beyond the confines of the prostatic capsule.

Radioimmunoassay of prostatic acid phosphatase (PAP) was reported in 1977 (3) and, since then, has been employed in increasingly large numbers of patients as a routine screening measure to discover early cases of prostatic carcinoma.

An elevated PAP in an individual whose prostate is clinically benign has become an increasingly common occurrence, and confronts the urologist with a difficult problem. Since there is no specific area to be biopsied, random biopsies are necessary, and, if these do not reveal the presence of carcinoma, the patient and physician are no better informed than they were prior to the procedure. It would seem advisable to follow these patients and perform a biopsy only when a suspicious area is present in the prostate.

Needle biopsy of the prostate can be performed either through the perineum or through the rectum. The procedure is usually innocuous, but complications such as sepsis and bleeding have occurred. In rare instances, seeding of prostatic cancer has occurred along the needle tract. Because of this, a perineal biopsy is preferable to the transrectal approach. The accuracy of a needle biopsy is approximately 90%. In those instances in which a truly operable carcinoma is thought to be present and a negative biopsy is obtained, surgical exposure of the prostate should be performed and an open biopsy made.

Cancer of the prostate has been clinically staged as follows:

STAGE A: Microscopic focus of cancer in less than three microscopic foci in TUR chips or specimen obtained at open enucleation. This is the so-called occult carcinoma which is discovered during the course of a simple prostatectomy performed for obstructive symptoms caused by benign hyperplasia of the prostate.

STAGE B-1: A small nodule less than 2 cm in diameter surrounded by normal tissue.

STAGE B-2: Extensive involvement, but not extending beyond the capsule.

STAGE C: Local extension.

STAGE D: Metastatic disease.

The clinical appearance and percentage of the various stages are as follows:

STAGE A: Bladder neck obstruction requiring surgery for benign hyperplasia (5–10%).

STAGE B: Incidental finding on routine physical examination in an otherwise asymptomatic individual (5–10%).

STAGE C: Obstructive symptoms, backache, sciatic pain (45%).

STAGE D: Obstructive symptoms, symptoms secondary to metastatic disease (35%).

There is perhaps no other disease in which more controversy exists as to treatment in its various stages, as carcinoma of the prostate. The scope of this paper precludes an indepth discussion of the merits of each position, and an attempt will be made to summarize the most widely accepted positions for each stage.

STAGE A: Most clinicians feel that this is a relatively benign condition and requires no treatment except for close observation. Other studies have shown a high degree of malignancy in these individuals and total prostatectomy has therefore been recommended. Intermediate positions are followed by others and radiotherapy or endocrine manipulation has been advised as further therapy.

STAGE B-1: Radical prostatectomy (with or without lymphadenectomy), or radiotherapy.

STAGE B-2: Radiotherapy or total prostatectomy.

STAGE C: Radiotherapy (external beam or interstitial implantation).

Observation.

Endocrine manipulation.

STAGE D: Endocrine manipulation.

Opinion is divided as to the proper management of patients with obstructive symptoms. Some urologists proceed with immediate transurethral resection. Because tumor cells can be demonstrated in

the peripheral circulation of patients undergoing transurethral resection, others prefer to treat these patients either by orchiectomy or estrogens.

Recent studies seem to indicate that estrogen increases the risk of cardiovascular and pulmonary complications in the aged, and therefore it would appear to be more prudent to perform orchidectomy in those patients in whom hormonal manipulation is necessary.

Painful gynecomastia may result from estrogen therapy. It has been suggested that these patients have their breasts irradiated prior to therapy. This, however, has not been uniformly successful in preventing this complication.

Radical prostatectomy should be advised only to those patients in whom no evidence of metastatic disease exists and in whom there is at least a ten-year life expectancy.

Pelvic lymphadenectomy has revealed a significant incidence of silent nodal disease in five of 21 patients with B-1 lesions, three of eight with B-2 lesions, and five of 18 with stage C lesions (4).

These studies would indicate that either lymph-

adenectomy or at least lymphangiography with Chiba needle biopsy of suspicious nodes should be performed before radical prostatectomy is advised.

Summary

A cursory review of current concepts of management of benign hyperplasia and malignant diseases of the prostate is presented.

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Surgery in the Elderly: Attitudes and Facts

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At the present time in the United States, eight to nine percent of the population has reached the age of 65 or older. This statistic reflects approximately 20 million people. Projections by the United States Census Bureau indicate that by the year 2000, some 30 million people (representing 10%-12% of the population) will be in this steadily expanding age group (1). Consequently, all aspects of senescence are being studied.

Geriatric medicine and surgery have developed as fields of specialized interest and knowledge over the past 20 years. This has been in direct response to the observation that older individuals, by comparison with others, have a disproportionately large share of medical and surgical problems. Patterns of hospital admissions confirm this well known clinical fact.

In 1968, Dr. John Powers who edited one of the first modern textbooks on surgery in the elderly patient stated that "surgery of the aged is different" (2). Over the past ten to 20 years, those of us involved in caring for this special segment of the population, by virtue of interests and because of a unique association with the Jewish Home and Hospital for the Aged in New York City, have come to understand this. To deal with the medical and surgical problems of elderly people intelligently and wisely, it becomes necessary to understand something about the aging process itself.

Fifty years ago, the eminent surgeon Alexis Carrell, following his experimentation with chick fibroblasts grown in vitro, came to the conclusion that animal cells that were removed from the regulatory mechanisms of the body might possess the property

of being "immortal" (3). The earliest proposal about the causes of aging suggested that circumstances being less than ideal, aging was the wearing out of certain parts of function of the human body. More recently in the 50's and 60's, Hayflick discovered that normal human cells growing in tissue culture were not "immortal." They would eventually degenerate and die after many generations of reproduction. It usually appeared that such cells have a capacity to replicate themselves about fifty times and then no more. Cells taken from older people duplicate themselves fewer times than those taken from embryos or the very young (4).

At the current time, most scientists involved in this area of research see little prospect of altering the aging process altogether, or even making major extension to human life which probably has a maximum of around 100 years. Only abnormal cells seem to resist death or at least they do until the death of the host organism (5).

The role of the physician treating elderly patients then becomes clear. He or she must intervene and deal in those pathologic processes that may appear during the finite time of man's expected life span. It must be remembered that even at advanced ages there are significant expected survival times (Table I) (6). It is probable that expected survival times have improved since the report of 1967. We now approach those areas of understanding that make

TABLE I
Average Number of Years of Life Remaining (U.S. Vital Statistics, 1967)

Age	Years Remaining for Males	Years Remaining for Females
At birth	67.0	74.2
60	16.0	20.2
65	13.0	16.4
70	10.4	13.0
75	8.3	10.0
80	6.4	7.3

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surgery of the elderly special and different. The technical aspects of an operation do not change when performing surgery on a younger or older person. The maximum skill of a surgeon is demanded whenever he approaches the operating table. What does change is the patient. While every patient being taken to surgery from the preoperative evaluation to the rehabilitative phase always combines the efforts of a number of physicians, it is critical for the surgeon operating upon the elderly to have an inherent understanding of the limitations of his patient. He must recognize the pitfalls and hazards, and have at his instant command a variety of possible alternative solutions for problems presented at the operating table. He must know when to be more aggressive or less aggressive to achieve the optimal result.

Therefore, the first bodies of knowledge with which a surgeon must have a working understanding are those biologic and physiologic changes that normally occur with aging. The graph in Figure 1 (7) is self-explanatory and extremely important.

It should come as no surprise that the decline in the various functional capacities is most profound in the realm of pulmonary function, with the renal and cardiovascular aspects close behind.

The multiple pathologic processes that are fre-

quently seen in addition to the normal biologic aging processes necessitate an enormous difference in attitudes and understanding for the aged patient. Approximately 75% of patients over the age of 70 have one or more associated diseases.

Preoperative Preparation of the Patient

Although efforts have been made to categorize the risk factors or operability of the elderly patient, no formula exists that is both accurate and clinically feasible. Schemes have been devised involving multiple types of preoperative testing, but they themselves often turn out to be an impossible strain for many of the elderly. We are left with a basic check list in the preparation of the older patient.

First and foremost, in both emergency and elective surgery, the blood volume and red cell mass must be brought towards normal levels. It is then a constant observation that the elderly often have a significant anemia. Dehydration, which occurs quite rapidly in the elderly patient, will often cause a deceptive elevation in hemoglobin or hematocrit, masking an underlying anemia that becomes manifest when the dehydration is corrected.

Secondly, since pulmonary complications rank among the most troublesome postoperatively, any

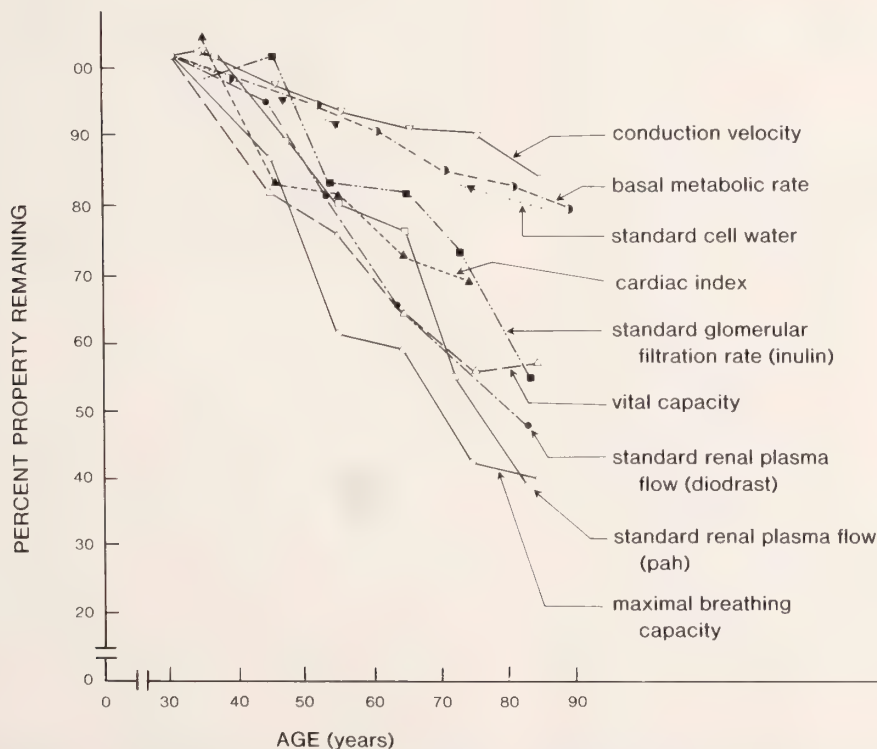


FIG. 1. Curves representing the loss of efficiency of several human physiologic functions in relation to age. (Kohn, R. R.: *J. Chronic Dis.*, 16:5-21, 1963).

preoperative pulmonary infection must be dealt with vigorously. While a full battery of pulmonary function tests is always useful, the simple measurement of forced expiratory volume or the simple ability to extinguish a match are more useful. If the patient fails either of these two latter tests, serious trouble can be anticipated. All but the most life-saving surgery should be deferred.

The renal status is probably best determined by the measurement of blood urea nitrogen, creatinine, and the ability of the urine to concentrate. A low fixed specific gravity is truly an ominous sign that the patient will do poorly with even the slightest degree of hypotension or anoxemia. Elevated blood urea nitrogen may represent dehydration and sepsis, and can be expected to return towards normal if these conditions are corrected.

Cardiac status: Physicians are aware of the high mortality rate associated with any surgery performed within three months of a myocardial infarction. Certainly, elective surgery should be deferred until six months have elapsed. Nevertheless, if there are indications to perform truly life-saving emergency surgery within this time limit it should be done. Uncontrolled congestive heart failure and arrhythmias are contra-indications for surgery. Even these can be controlled in hours or days if emergency surgery is indicated. Drugs, cardioversion, and temporary pacemakers have all been used rapidly to convert an unstable cardiac condition to a relatively stable one.

Nutritional status must be considered in the preoperative evaluation. The surgeon is primarily concerned with the processes of wound healing and inflammatory responses that should normally occur. There is no question that surgical wounds do heal in the aged despite starvation, anemia, uremia, diabetes or generalized decreased organ function. Yet, most surgeons believe that wounds heal more slowly and with a greater number of complications under these circumstances. The causes of nutritional deficiency in the elder are diverse and range from chronic illnesses, lack of information about proper diet, poor dentition, and often simply lack of money to purchase proper food. It would almost seem that the socioeconomic factors often exceed the medical reasons. Often attempts must be made to correct these deficiencies in a short time. It can also be unrealistic to believe that the deficiencies can be corrected in a matter of days or even weeks when they are the cumulative products of months or even years of inadequate intake. The use of parenteral alimentation, elemental diets, and tube feedings, however, have all found an important

place in achieving at least a steady state in short-term situations of urgency.

The liver probably remains an under-rated cause of potential problems in the elderly surgical patient. Postoperative derangement of hepatic function seems to be disproportionately high. Since the liver is involved in so many body syntheses, degradation processes, and metabolic controls, it is very hard to pinpoint specific areas of compromise except in the most florid situations. Patients with cirrhosis, acute or chronic active hepatitis, and coagulopathies represent contra-indication of all but the most life-threatening surgical situations.

Cerebral dysfunction is something with which the surgeon, perhaps, is mistakenly assumed to be unconcerned. We feel strongly that this is one of the most critical areas for him to observe. There are moral and ethical issues involved as well as the medical problem. However, for the moment we will defer the discussion of the severely impaired elderly patient who has a surgically treatable disease. A much more common situation that is appreciated after significant experience with elderly patients is the transient derangement that can take place. Stress, whether caused by fever, dehydration, trauma, sepsis, pain or even the prospect of imminent surgery is capable of producing marked temporary mental disorder. Disorientation, hallucination, agitation and depression can develop a new or make worse any pre-existing situation. The surgeon must know this for two reasons. First, he has to reassure himself that he is doing justifiable surgery. Secondly, he can advise the family that there will usually be a return to the pre-existing mental status.

Attempts to correlate the adverse cerebral effects of stress with hypotension, anesthesia, blood gas alterations, and metabolic changes have been made periodically (8-10). All seem to contribute. None is the complete answer.

Role of Anesthesia

The success of surgery in the aged is inextricable from the success of anesthesia in these patients. At The Mount Sinai Hospital, we are fortunate in having a group of anesthesiologists who are interested as well as expert in dealing with these patients. In 1977 Miller, Marlar and Silvay reviewed the departmental records from 1970 to 1975 of patients over 90 years of age undergoing surgical procedures. There were no fatalities in the operating room or the recovery room in 147 patients undergoing 171 surgical procedures (11). It is becoming increasingly recognized that the immediate post-

operative period is the most hazardous for the elderly patient. Postoperatively, many of the patients are literally too exhausted to breathe or ventilate adequately after major surgery. Prolonged ventilatory support and careful monitoring of blood gas levels, acid-base balance, fluid and electrolyte balance, etc. are critical factors in the postsurgical care. The availability of trained personnel in the recovery room, and a backup intensive care unit capable of providing prolonged ventilatory support and monitoring often extending to hours or days, is critical to the successful treatment of these geriatric surgical patients. Probably, major surgical procedures in the very elderly age groups should not be undertaken unless these facilities and personnel are expert and available.

Statistical Review of Major Colorectal Surgery

Since we are now advocating that necessary surgery can be performed regardless of age—what are the results? In October 1979, one of the authors was called upon to treat a lady with an obstructing carcinoma of the right colon. She was one hundred-years old. After rapid preliminary work up and resuscitation were carried out, an emergency right hemicolectomy was performed successfully. The patient was returned to her nursing home approximately two weeks after the surgery.

This prompted a retrospective review by one of the authors of 100 consecutive major unselected colorectal cases in patients 65 or over. It is presumed that these cases represent a typical array of the types of such cases that would be seen by a general surgeon dealing with diseases of the elderly. Lesser procedures and pathology such as hemorrhoids and perirectal infections have been excluded from this series.

Results

Table II shows the distribution by age and sex which supports the figures that more women live into older age than men. The 11% mortality rate includes emergencies as well as elective cases. Considering the magnitude of surgery performed, these figures compare favorably with other series reported in the literature that deal with procedures of similar magnitude and in similar age groups. Mortality figures reported seem to range between 10% to 25% (12–15).

Table III illustrates, as expected, that the vast bulk of colonic surgery is for carcinoma. The majority of operations performed were resections, with only a few bypass procedures being carried out for unresectable lesions.

It is noted that four cases of stercoral perforation of the colon are present and these represent an entity seen mostly in the aged, and one that should be preventable. The author has reported previously on these patients (16).

Also, all of the eight cases of diverticular disease surgery were performed either for obstruction or, more likely, perforation whether localized or generalized. No surgery was performed for massive hemorrhage in this condition. Inflammatory diseases were conspicuous by their absence or low number of cases.

In surgical lesions of the rectum, carcinoma was the most common major disorder (Table IV). Of the carcinoma cases, five patients underwent abdominoperineal resection and four were treated by either local resection or fulguration. The oldest

TABLE II

*100 Consecutive Cases of Major Colorectal Diseases Treated Surgically in Patients 65 and over.
December 1973–October 1979*

Age	Male	Female	Mortality
100+	—	1	0
95–99	—	—	—
90–94	1	4	1
85–89	1	8	2
80–84	5	14	0
75–79	4	18	5
70–74	8	17	3
65–69	11	8	0
Total	30	70	11

TABLE III

100 Consecutive Cases Pathology–Colon

Lesion	Number of Cases
Carcinoma	
right colon (30)	69
left colon (39)	
Villous adenoma	2
Diverticular disease	8
Sterocoral perforation	4
Colostomy prolapse	2
Volvulus	2
Granulomatous disease	1
Traumatic perforation (iatrogenic)	1

TABLE IV

100 Consecutive Cases Pathology–Rectum

Lesion	Number of Cases
Carcinoma	9
Villous adenoma	1
Rectal prolapse	1

patient undergoing abdominoperineal resection was a woman 82-years old, who, ultimately, did not manage her colostomy too well. Another patient was managed well by repeated fulgurations beginning at age 95 until her death at age 102.

Table V represents the usual marked discrepancy in mortality rates that occur when dealing with elective versus emergency surgery in this or any age group. Of the six deaths occurring in the emergency series, three were associated with stercoral perforation. This, indeed, represents a highly lethal condition.

The overall mortality for the number of actual procedures performed was 8.7% (Table VI). There appears to be no appreciable difference in mortality rate between the patients requiring one operation or those requiring two procedures. The mortality related to the type of disease and its severity rather than to the number of procedures or the age of the patient.

Table VII shows the cause of death in the six emergency cases. This entity of stercoral perforation represents exaggerated fecal impaction probably with pressure necrosis of the wall of the bowel with sudden rupture and massive fecal peritonitis. In only one case was there a specific disease entity as the underlying pathophysiologic cause, namely

scleroderma of the bowel. In other cases and in those cases previously reported, there was high grade impaction caused by chronic constipation in the absence of associated bowel pathology. The group of patients seen with this entity are the elderly, the institutionalized, and the mentally impaired.

The two other patients in this group of cases represent advanced malignant disease with extensive carcinomatosis in each case. Again we emphasize that mortality in these patients is related to the severity of the disease rather than any other factor.

Table VIII. In the mortalities for the elective cases, severe associated disease in cases one and five accounted for the death—weeks after the colonic surgery had been completed and the patient was functional (Table VIII). However, in each case, the patient never left the hospital and succumbed from secondary diseases even though well beyond the 30-day time period included in surgical mortality calculations.

Moral, Ethical and Other Considerations

Thus far, we have presented only the idea that the surgeon must have increasing knowledge about

TABLE V

100 Consecutive Cases Initial Presentation of Patients

Emergency		Mortality
21		6
	Colon obstruction	9
	Stercoral perforation	4
	Diverticulitis with perforation	5
	Prolapsed colostomy	1
	Volvulus	1
	Traumatic perforation	1
Elective		5
79		

TABLE VII

100 Consecutive Cases Analysis of Mortalities

EMERGENCY CASES
1. S.S. (F-75) stercoral perforation; scleroderma; peritonitis, shock.
2. L.F. (F-79) perforated diverticulitis; metastatic Ca breast; sepsis.
3. S.B. (F-87) stercoral perforation; sepsis, pneumonia.
4. V.J. (F-75) obstructing Ca transverse colon with metastases; carcinomatosis.
5. C.M. (F-73) obstructing Ca descending colon with metastases; carcinomatosis.
6. S.S. (M-90) stercoral perforation; sepsis, shock. (A)*

* (A) autopsy performed.

TABLE VI

100 Consecutive Cases Patients Requiring Multiple Operations

Number of Patients	Procedures Per Patient	Number of Procedures	Mortality
80	1	80	9
16	2	32	2
3	3	9	0
1	4	4	0
Total Patients		Total Procedures	
100		126	
Mortality		Mortality	
11%		8.7%	

TABLE VIII

100 Consecutive Cases Analysis of Mortalities

ELECTIVE CASES

1. M.P. (F-77) 2nd stage colon resection for Ca; kidney, congestive heart failure, renal failure; arrhythmia.
2. I.F. (F-86) Left colon resection for Ca; prolapsed small bowel through drain site; sepsis, shock. (A)*.
3. H.J. (F-72) Prolapsed colostomy requiring laparotomy; obesity, hypertension, previous CVA; CVA. (A)*.
4. B.P. (F-77) 2nd stage colon resection for Ca involving spleen; empyema. (A)*.
5. G.Z. (M-72) Colon resection for Ca with imminent obstruction; glioblastoma multiforme; inanition, convulsions. (A)*.

* (A) autopsy performed.

the biologic processes of aging. It is also necessary that the surgeon be familiar with the social and psychologic effects of aging and its effect on the clinical presentation of the illness. One of the most difficult problems he must face when dealing with the elderly patient is, namely, should he operate at all. For example, is an operation necessary to relieve obstructive jaundice from whatever cause in a patient in the 80's with severe brain dysfunction because of associated disease? Does one prolong life at any cost?

The geriatric surgeon must be extremely familiar with the concepts of rehabilitation medicine. Although surgical wounds heal in the elderly, there is no question that the total functioning organism responds differently at the age of 85 than at 25. It must be explained with many reassurances to both patient and family that the patient will get well, but that it will take longer.

An educated awareness of the biologic, sociologic and psychologic aspects of the geriatric population becomes a necessary therapeutic tool in caring for the elderly. The general surgeon, whose training and inclination often have short term, doctor-patient relationships, must realize that geriatric surgery requires a unique approach and attitude in the same way that pediatric medicine and surgery requires a special feeling for the patients at the other end of the age spectrum. This is particularly true in dealing with the increasingly large number of patients who may have outlived their friends and whose families may be distant or otherwise unavailable. The feelings of helplessness in these patients may be quite acute. It is well worth the time and effort of the surgeon to attempt to dispel some of the apprehension that is normal in patients facing surgery, but often compounded in the elderly by their life circumstances.

Two of the most common statements heard when

confronting a preoperative patient who is elderly are "I am too old to be operated on," and "I have lived my life and I am ready to die." The fears inherent in the first statement usually can be alleviated by a thoughtful discussion with both patient and family, pointing out exactly what is involved, what is to be expected, and what the results can be. The second statement of being "ready to die" is usually far from the truth, unless the patient is experiencing severe chronic pain or depression. Both of these conditions can be relieved by the timely and appropriate interventions of the attending physician and surgeon.

Essentially, geriatric surgery is something more than surgery of the elderly. Geriatric surgery exceeds the sum of its technical and scientific aspects and becomes an art as well as a science when practiced well.

Conclusion

Chronologic age should never be a deterrent to necessary surgery. What makes this surgery unique and demanding is that there is very little margin for error because of diminished organ function in the elderly patient. These limitations must be fully appreciated by the physicians undertaking the care of these patients. In addition, the attitudes of the patient, the family, and the physicians are extremely important in restoring the elderly patient to his normal state of health and capacity.

A series of one hundred major colorectal cases has been presented to illustrate that surgical risks in the older age groups are quite acceptable and will probably be improved. We anticipate that further education and research will make surgery in the aged less hazardous.

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Vascular Surgery in the Aged

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Abstract

The elderly patient has characteristics that require special consideration in the determination of how to best manage the abnormalities of the blood vessels. Because of the inherent increased operative risk in the aged patient, it is particularly important for the vascular surgeon to utilize those procedures that can be done expeditiously and are best suited to deal with the problem in the context of a limited life expectancy.

Introduction

During the past twenty-five years, rapid strides have been made in the surgical management of abnormalities of the blood vessels. Many of these advances have been made feasible by the parallel improvements in the anesthetic and medical management of the relatively poor risk patient. It is the integration of the services of the anesthetist, internist, and vascular surgeon that makes possible the salvage of limbs and lives that would have been considered impossible just a few years ago. Preoperative, intraoperative, and postoperative care require critical application of the principles of these three disciplines.

Preoperative

The general appearance of the patient is frequently a good guide to the advisability of strong consideration for surgery. A nonresponsive elderly patient with a severe organic mental syndrome is obviously not a suitable candidate for major surgery in the hope of prolonging that vegetative form of life. Likewise, major arterial reconstruction to improve circulation to the legs is of little use in the patient who is confined to bed or to a wheelchair. Severe cardiac, pulmonary, and renal disease may also offer serious contraindications to major vascular surgery. However, age alone should not be a contraindication since major vascular procedures have been successfully performed on patients well

into their nineties. This is especially true now that relatively recent advances have made possible satisfactory revascularization with minimal disturbances to the patients' homeostatic mechanisms. These advances have been both in the anesthetic management and in the surgical procedure.

Anesthetic Management

In the elderly patient, local anesthesia supplemented with intravenous agents has made possible the revascularization of the extremities with minimal anesthetic risk. The employment of axillary-femoral and femoral-femoral bypass under local anesthesia has obviated the need for laparotomy to bypass iliac artery obstruction. Likewise, saddle embolus can be successfully treated with the use of a Fogarty catheter introduced via the femoral artery under local anesthesia. Most emboli to the extremities can now be treated with a Fogarty catheter without the use of general anesthesia. Even femoropopliteal grafts may be inserted under local anesthesia, although intravenous supplement is necessary, and the procedure may be difficult and tedious. Abdominal aortic aneurysm and visceral emboli are practically the only vascular lesions in the elderly requiring laparotomy and general anesthesia. Postoperatively, the use of modern ventilatory machines in combination with the endotracheal tube has salvaged many elderly patients who were unable to ventilate adequately during the early postoperative period.

Surgical Management

In the past two decades, there has been a gradual evolution of the vascular procedures employed in

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the elderly. The following anatomic categories are accompanied by a discussion of the preferable surgical procedure.

Extracranial Cerebrovascular Insufficiency. Aged patients with cerebral symptoms are infrequently suitable candidates for revascularization of their carotid arteries because of associated intracranial disease. However, in otherwise good surgical candidates, a solitary internal carotid stenosis or ulceration (which is symptomatic) can be repaired under local or general anesthesia. General anesthesia is usually preferred because cooperation of the patient is mandatory when this type of surgery is done under local anesthesia.

Abdominal Aorta and Its Visceral Branches. Occlusive disease of the infrarenal abdominal aorta can be safely accomplished by utilizing an axillary-femoral or axillary-bifemoral graft under local or general anesthesia. Laparotomy need not be done and the trauma to the patient is minimal, although the long-term results of axillary-femoral grafting are not as good as with aortofemoral grafting. The results in these elderly patients with a limited life expectancy have been gratifying.

Abdominal Aortic Aneurysms are severe threats to life. It is now well documented that some small aneurysms rupture, but in general an aneurysm smaller than 5 cm in diameter can safely be kept under observation with periodic sonography (1). With this very accurate modality, an enlargement of even one cm can be readily detected and appropriate measures can then be taken. Life expectancy and operative mortality rates must be carefully considered when determining the advisability of aneurysm excision. The average life expectancy at age 75 is approximately ten years and, at age 85, it is approximately six years (2).

The mortality rate of abdominal aneurysmectomy in asymptomatic patients over the age of 75 was 14% in the hands of Esselstyn and his colleagues (3). Coupled with the above mentioned statistics, the incidence of rupture in untreated cases must be considered. This rate was 63% (within ten years) in the oft-quoted series of Estes (4), and 44% in the series of Schatz (5). It may be said that there is reasonably good indication for aortic aneurysmectomy in an elderly patient with an aneurysm larger than 5 cm. in diameter, and whose life expectancy is at least two to three years.

Visceral Arteries. Embolus to a visceral artery is indication for laparotomy and attempt at direct revascularization of the vessel. Unfortunately, satisfactory results from superior mesenteric artery embolectomy are extremely rare. Somewhat better

results are obtained with renal artery embolectomy, but surgery is frequently not necessary for the survival of such a patient.

Saddle Embolus can be successfully treated under local anesthesia and with a Fogarty catheter introduced from the femoral artery.

Iliac Artery Obstruction, if unilateral, is readily treated with a femoral-femoral graft under local or general anesthesia. Axillary-femoral bypass, although not as reliable as femorofemoral bypass may be used for unilateral or bilateral iliac artery obstruction under local or general anesthesia. Placement of the axillary-femoral graft in such a manner so as to minimize the compression of it in a patient who spends the major portion of life in bed is extremely important.

Superficial Femoral Artery Obstruction causing ischemia of the distal leg is probably the most frequently encountered indication for arterial reconstruction in the aged. Very often the surgeon is confronted with an elderly patient who has a small area of gangrene on a toe or who has an obviously ischemic foot with rest pain. The decision must then be made whether to perform angiography and possibly bypass, or to proceed with leg amputation. Needless to say, the angiogram may be of great help in deciding on an attempt at revascularization. However, the patient frequently is a bedridden patient with an organic mental syndrome for whom the limb is little more than a cosmetic appendage. If this be the case, the risk of revascularization is hardly justified by the possible gain. If, however, the patient is a reasonable operative risk and appears to be able to benefit from vascular reconstruction, operation may be resorted to. It is then most important to perform the surgery expeditiously. The availability of the Goretex prosthetic graft for the past few years has made it possible for reasonable results to be obtained with an operating time of about seventy-five minutes. Since the depth of anesthesia necessary for this procedure is not great, the anesthetic risk is acceptable.

If such a short operative time is contraindicated because of a debilitated medical condition, intraluminal angioplasty as devised by Gruntzig may be employed (6). This technique is only suitable for localized lesions that are not obstructing, and the long-term results are not yet known. However, since the risk of anesthesia is avoided, the procedure may have an application in a very limited number of patients.

When none of the above procedures can satisfactorily salvage a limb, consideration must be given to amputation. Amputations of toes or through the

metatarsals are rarely possible in these elderly patients because their peripheral circulation and skin vitality usually are not adequate for healing. Below knee amputations should be the choice in patients who are adjudged in sufficient possession of their faculties (both mental and physical) to allow them to use a prosthesis, or at least to use the stump below the knee as a fulcrum for turning in bed (7). If these criteria cannot be met, above knee amputation should be done forthwith even though the circulation is adequate to maintain a below knee stump. Below knee stumps in such patients are subject to constant trauma and the frequently encountered flexion contracture of the knee results in ulceration and pain in the distal end of the stump requiring eventual above knee amputation. The multiple operations on the legs make these patients especially prone to pulmonary emboli. In the patient who is confined to a nursing home, prompt above knee amputation is the procedure of choice in the majority of cases. A mortality rate of 20%-40% should be expected in such amputations (8). This risk must be explained to the concerned children so they may intelligently share in the decision making in regard to operation on their parent. Occasionally, the decision to withhold amputation is made. This is usually done to spare the patient the loss of a limb prior to death which is expected to occur within a short time, usually thought to be a few days. Unfortunately, the situation rarely resolves itself in such a "favorable" manner. The usual course is a prolonged one of several weeks in spite of the patient appearing to be practically moribund at the time of making the decision. Pain becomes prominent and requires large amounts of narcotics that must be administered carefully to control the pain and yet not excessively so it won't

be considered an overdose and a practice of euthanasia. Difficult nursing problems arise because of the gangrene and also because of severe bed sores. For these reasons, when the decision has been made to allow a patient to die because of a gangrenous limb that will be amputated, all involved staff must be apprised so that only the most minimal of support measures be continued. Antibiotics, daily fluid requirements by intravenous route, and costly laboratory studies are to be avoided. As discussed above, the family must participate in and actually support these decisions.

Last scene of all,
That ends this strange, eventful history.
Is second childishness, and mere oblivion
Sans teeth, sans eyes, sans taste, sans everything.

As You Like It—Shakespeare

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Ambulatory Management of Lower Extremity Venous Disease in the Aged

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Since venous stasis invariably becomes worse with the passage of time, its complications are seen frequently in the aged. Neither varices nor edema usually causes significant symptoms until complications ensue. Patients with varicose veins can develop superficial phlebitis, hemorrhage from a ruptured vein, or ankle ulceration; whereas brownish discoloration, flare-up of a deep vein phlebitis, or ankle ulceration are common with uncontrolled chronic postphlebotic edema. In the treatment of these conditions in elderly patients, excessive use of drugs and prolonged immobilization should be avoided.

Great strides have been made in recent years in the field of vascular surgery. Not only have highly innovative procedures been devised, but the period of hospitalization has been gradually decreased so that there is minimal bed rest. This is particularly important in older individuals, who have a relatively high susceptibility to venous thrombosis. Despite this recent progress, the incidence of phlebitis and pulmonary embolization has remained essentially unchanged during the past decade. One to two percent of all surgical patients develop thrombophlebitis postoperatively and 0.5 percent experience pulmonary emboli (1). Although the exact mechanism for the development of thromboembolic phenomena remains obscure, platelet adhesiveness, stasis resulting from inactivity and dehydration may all be incriminating factors, especially in the aged. Early ambulation has long been recognized as a vital factor in minimizing postoperative complications but it is only in recent years that ambulatory surgery has gained universal acceptance. This re-

port describes those lower extremity venous problems that occur in elderly patients and that can be managed effectively on a fully ambulatory basis. Even when surgery and anesthesia become mandatory, these techniques have greatly minimized surgical and postoperative complications.

Thrombophlebitis

Superficial phlebitis, in contradistinction to the deep variety, is rarely, if ever, responsible for pulmonary emboli. These patients are frequently put to bed and given wet compresses, antibiotics and other medication. Not only is this immobilization unnecessary, but it is also conducive to the more dangerous deep vein phlebitis. In the average patient, an elastic bandage from the toes to the knee will relieve discomfort and allow him to work and carry out all of his usual activities while the inflammation subsides. Antibiotics are not indicated because the inflammation is nonbacterial. Spontaneous recovery usually occurs without medication. If the inflammation is unusually severe, anti-inflammatory agents, such as Butazolidin®, are helpful. This is a potent drug and should be used with caution. It is usually sufficient to give 100 mgs T.I.D. for three to four days. If the phlebitis extends upward along the long saphenous vein and approaches the upper thigh or groin, a simple high ligation, under local anesthesia, would be indicated to prevent extension of the inflammation into the femoral vein.

If deep vein phlebitis develops, one week of bed rest and anticoagulants are indicated. The patient can then become ambulatory since all loose thrombi should have become fixed to the vein wall by that time. Anticoagulants should be continued for another six weeks. The best way to prevent additional episodes of phlebitis is for the patient to remain fully active. If pulmonary emboli develop in spite of

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anticoagulants or if emboli occur and the use of anticoagulants is contraindicated because of other associated conditions, such as a duodenal ulcer, surgery is indicated to prevent additional emboli.

It is unwise to ligate the superficial femoral vein because it does not completely protect against pulmonary emboli. A ligation of the common femoral vein (proximal to the entrance of the saphenous vein) is not indicated because many patients will develop significant complications. Common femoral vein ligations were performed to prevent pulmonary emboli in 103 extremities (2). In only ten instances were there no sequelae whatever. In the remaining 93 extremities, there was some evidence of venous stasis. This was manifested by minimal edema in some cases, but other patients developed marked edema, pain, pigmentation, and intermittent ulceration of the leg. In patients with marked venous stasis, the symptoms and physical signs of venous impairment often became apparent immediately. However, postphlebotic and postligation effects often appeared for the first time several years or longer after the active phlebitis or vein ligation. Once symptoms started, they usually became gradually worse, especially if the edema was not well-controlled.

If conservative measures fail and surgery becomes mandatory to prevent pulmonary emboli, the inferior vena cava site is optimal since there is good collateral circulation at this level. Many ingenious clips and umbrellas have been devised recently to prevent significant-sized thrombi from ascending to

the lungs. The goal of surgery is the elimination of emboli without completely occluding the inferior vena cava.

Stasis Eczema and Ulceration

Venous stasis can be caused by either varicose veins or edema. Varicose veins do not produce significant edema, which is the result of deep vein insufficiency. Despite extensive research and improved clinical care in recent years, there still remains a large group of patients with clinically significant venous stasis. This is most commonly the result of a previous phlebitis. The postphlebotic edema often first appears several years after the acute episode and many patients are not even aware that they had phlebitis. The edema starts insidiously and the patient is usually not aware of it because it does not cause any significant symptoms. The lower leg region gradually becomes discolored and, with the progression of time, eczema or ulceration develops because of the uncontrolled venous stasis. The postphlebotic syndrome is especially prevalent among the aged.

When eczema, due to varicose veins or chronic edema, first appears, the usual picture is one of neglected venous stasis with chronic congestion and discoloration of the skin surrounding the medial malleolus (Fig. 1). The eczema usually starts in the malleolar region and then spreads to adjacent areas. By the time the patient is seen, many ointments and other local medication have been used, fre-



FIG. 1. Stasis eczema caused by chronic venous insufficiency. If all venous stasis is effectively eliminated with pressure bandages, healing invariably takes place on an ambulatory basis. Local or systemic treatment is unnecessary.

quently resulting in a generalized allergic skin eruption. The crux of effective treatment is the elimination of venous stasis. A gelatin paste bandage (Unna's boot), such as the Medicopaste Bandage (Graham—Field Surgical Co.), has proved effective for varicose eczema. This bandage will compress varices but does not exert sufficient pressure to eliminate edema. If there is marked deep vein insufficiency, as evidenced by significant edema, the boot should be applied early in the morning, before edema has developed. No local medication is used. The patient can be fully ambulatory. The boots are changed weekly. Occasionally, the skin is so irritable that the boot cannot be tolerated. This is especially common in diabetics. In these cases, compression of the varices is accomplished by means of rubber reinforced elastic bandages, that are worn during the day and removed at night.

A stasis ulcer is frequently precipitated by trauma but it is the venous stasis that delays healing. With the use of bed rest or other types of treatment, it is often possible to accomplish temporary healing of stasis ulceration but the ulcer usually recurs if the underlying cause is not corrected. There is no surgical or injection procedure that can effectively eliminate the edema due to deep vein insufficiency. Skin grafting and other elaborate surgical procedures can produce healing of an ulcer. However, the edema usually persists and the ulcer invariably recurs unless proper elastic supports are worn. Healing can be accomplished on a fully ambulatory basis with the use of compression bandages (3).

Stasis ulcers usually develop in the ankle region (Fig. 2). Most are caused by chronic edema, not varicose veins. Healing can be achieved by the use of compression firm enough to eliminate edema. Bed rest for about six weeks can also produce healing since it eliminates edema but this immobilization is especially undesirable in the aged. Although there are many ways to achieve adequate compression, medicated bandages (Primer by Edward Taylor) and elastic adhesive bandages (Ultra-plast by Edward Taylor) have proved simple to use and effective in the healing of stasis ulcers. The porous nature of the material minimizes maceration of the skin. The medicated bandage is applied first, from the toes up to the knee, and then the elastic adhesive bandage is applied firmly over the medicated bandage so that the adhesive does not come in contact with the skin (Fig. 3). This "boot" is changed at weekly intervals and healing of an average ulcer takes from five to eight weeks. No local or systemic treatment is used. The patient is advised to work and carry out all normal activities.



FIG. 2. Large stasis ulcer occurring in typical location, overlying lower medial portion of leg. Healing can usually be accomplished by using specially designed boots, applied at weekly intervals.

If it is not possible to use firm compression because of arterial insufficiency (Fig. 4), an Unna's paste boot is applied at weekly intervals. This bandage becomes firm after application but it will not produce sufficient pressure to eliminate edema already present. In these cases, the patient is advised to leave the ulcer exposed to the air while in bed at night. In the morning, when the edema usually has subsided, sterile gauze is applied over the ulcer and then a four inch rubber reinforced elastic bandage is applied from the toes to the knee. The patient should then report as soon thereafter as possible for the application of the Unna's boot. In this way, the boot will be applied to an edema-free leg and healing should take place. Sometimes an elderly patient cannot tolerate an Unna's boot. Since the basis of effective treatment is the elimination of venous stasis in the region of the ulcer, pressure is exerted by using gauze, then foam rubber over the ulcer, followed by firm wrapping of the leg with a rubberized elastic bandage during the day. The ulcer is left exposed to the air while in bed at night. Only in



FIG. 3. Application of pressure boot: The inner layer has already been applied from toes to knee and the elastic adhesive bandage is now being wrapped. If used sufficiently firm to eliminate venous insufficiency, this stasis ulceration should heal on ambulatory basis.

these cases is medication (a small amount of hydrocortisone and oxytetracycline) used, to prevent the bandage from sticking to the ulcer. If sufficient pressure is exerted, healing frequently takes place despite the arterial disease.

The patient is instructed to wear elastic bandages during the day indefinitely. If edema persists, the ulcer will recur. An elastic bandage is preferable to a rubber stocking since the stocking stretches after several weeks of use and is not adjustable. Men do not object to elastic bandages but many women prefer the stockings for cosmetic reasons. For this reason, female patients are advised to wear the bandage at home but to use a properly-fitted stocking when out-of-doors.

Management of Varicose Veins

Varices are one of mankind's most common afflictions. Man has not yet adjusted well to the upright position. Although the exact etiology of

varicose veins remains obscure, a familial predisposition is probably the most important cause. Basic abnormalities prevail in the vast majority of patients and if appropriate therapy is instituted, excellent results can be anticipated. If there is no significant valvular insufficiency, injection treatment (sclerotherapy) is effective. However, if there is very high retrograde pressure because of marked valvular incompetence, a ligation is essential in order to eliminate the source of the varices. Sclerotherapy is particularly useful in the aged because it can be performed as an ambulatory office procedure.

Injection Treatment

If there are unsightly spider veins in elderly patients, they can be camouflaged by using make-up. Treatment should only be instituted if the varices are very large or produce significant problems, such as recurring episodes of phlebitis. If the patient is



FIG. 4. Indolent stasis ulcer, complicated by associated arteriosclerotic occlusive disease. Pressure boots cannot be used because of arterial insufficiency. Local venous stasis can often be eliminated and healing achieved by using sponge rubber over the ulcer and then elastic bandages (not too tight because of the arterial disease) from toes to knee.

bed-ridden, active treatment should not be given. Injections should only be given to a fully ambulatory patient since there is a relatively high incidence of complications if he cannot walk and be normally active immediately following the injection. If large, symptom-producing varices are not eliminated, the patient is susceptible to thromboembolic disease, bleeding from ruptured varices, and ankle ulceration. These complications are especially difficult to manage in the aged because of frequently associated arterial disease and other systemic lesions.

The development of Sotradecol (Elkins-Sinn, Inc.) as a sclerosing agent has greatly improved the results of injection therapy. It is a sodium salt of tetradecyl sulfate and has a potent thrombogenic action. Allergic reactions or other side effects are rare. The solution is clear, nonviscid, has a low surface tension and is readily miscible with blood. A three percent solution is used for large varices, while the one percent concentration is employed for "spiders" and very small varicose veins. The technique suggested by Orbach (4) has proven effective. Dilution of the sclerosing solution by blood is minimized by introducing a small air bubble or foam into the lumen of the vein immediately preceding the injection. There is no danger of air embolism and the results reveal a high incidence of obliteration. Fegan (5) described a modified injection technique that involves continuous compression of the injected veins for six weeks.

The injection technique can be carried out in several different ways. The patient can stand erect while the needle is being introduced (the veins assume their greatest prominence in this position). As soon as the vein is punctured, the leg is raised to hip level to empty the vein. The injection (usually $\frac{1}{4}$ to $\frac{1}{2}$ cc) is given and the leg is then wrapped with an elastic bandage which is not disturbed for 48 hours. Some physicians inject veins with the patient lying down, some use tourniquets and some leave elastic bandages for as long as six weeks. The most important factor is the injection into an empty vein, followed by firm compression. Following injection treatment, the patient should remain fully active. He can immediately return to work and all normal activities. If the varices are scattered and there is no retrograde flow resulting from marked valvular insufficiency, injection treatment is very effective.

Surgical Treatment of Varicose Veins

Varicose veins should not be treated surgically in the aged for cosmetic reasons. If the patient's general condition is satisfactory and if the varices are large and cause recurring complications, such as

phlebitis, hemorrhage or ankle ulceration, surgery would be indicated. If the varices are large and cause significant complications, but the patient's medical condition contraindicates surgery, then elastic supports should be worn during the day to compress the varices and minimize symptoms and complications.

If surgery is indicated and there are large varicose veins due to marked valvular incompetence, a high ligation and stripping procedure are indicated to eliminate all valvular disease at its source. In the majority of patients, most venous incompetence originates at the saphenofemoral junction. In about seven percent of patients, the short saphenous vein is diseased. There may also be incompetent perforators. When stripping procedures are performed for varicose veins, it is customary to hospitalize the patient for three to seven days. The length of hospitalization has been gradually decreased so that at the present time a complete stripping procedure can be performed on an ambulatory basis.

The physical examination and all necessary laboratory tests are performed one week before surgery. The patient shaves the operative area and has nothing by mouth after dinner the night before surgery. The patient reports to the ambulatory room at 7:15 a.m. and the anesthetist reviews his medical history to determine the best type of anesthesia. As soon as the patient reaches the operating room his varices are marked out with Flomaster ink (6), an effective skin marker that does not come off when the leg is prepared with Betadine (R). The use of simple inspection and palpation has been found to be accurate in the detection of sites of valvular incompetence (7).

The operative procedure should include the following: 1) elimination of all venous incompetence at the source; 2) excision of as many residual vein segments as possible so that few, if any, postoperative injections are necessary; 3) the making of small incisions so that the cosmetic result is good, and 4) mobilization of the patient within a few hours postoperatively to minimize thromboembolic and other complications. Although a stripping procedure can be carried out under local anesthesia (8), it is more pleasant for the patient to have a general anesthesia. Spinal anesthesia is not advisable because the patient will be unable to walk within a few hours following operation. A combination of agents is required that will provide a smooth induction and a light plane of anesthesia, followed by a tranquil emergence free from nausea, vomiting or other undesirable sequelae. Furthermore, the patient has to be able to walk within a few hours after surgery and be able to go home that same afternoon. An effec-

tive anesthetic technique has been devised by Dr. David Stark and his associates at The Mount Sinai Hospital, New York City. For patients who have not had recent surgery, the requirements for this type of ambulatory surgery have been met very well by the use of a barbiturate-nitrous oxide-halothane sequence. For those patients who have had recent anesthesia, halothane has been avoided. Intubation is used if the patient has to be turned to the prone position for a stripping of the short saphenous vein. The electrocardiogram, blood pressure and axillary temperature are monitored in the usual way.

The diseased long saphenous vein is first ligated flush with the femoral vein. Unless this is performed meticulously, recurrence of varices is inevitable because of missed branches very close to the femoral vein. After the high ligation the long saphenous vein is exposed just below the medial malleolus. It is divided and all adjacent branches are ligated. An intraluminal stripper is then passed upward and the tip brought out through the inguinal incision. If there are large varicose tributaries of the saphenous system, as many as possible are excised before the actual stripping is performed. With accurate skin marking, the incisions can be very small. Incompetent perforators are ligated flush with the deep system. The head of the operating table is lowered and then the long saphenous vein is stripped out slowly. If the short saphenous vein is diseased, the patient is turned over to the prone position and this vein is also stripped out. There is very little bleeding during surgery. The leg and thigh are wrapped with elastic supports from the toes to the upper thigh and the patient transferred back to the ambulatory room. About three hours after surgery, the patient walks with assistance. At about 2 p.m. he attempts to walk by himself and gradually becomes fully active. After walking on his own for at least one hour he can be discharged. No one is permitted to leave the hospital unless someone is available to take him home. Practically all of the patients have been discharged from the hospital between 2 and 4 p.m. If they are unable to walk normally by that time, they are advised to wait another hour or two until the residual effects of the anesthesia have disappeared. All patients are advised to rest in bed after returning home that night but to become normally active the next morning. Since no preoperative medication is used and there is practically no bed rest, the aged individual is much less likely to develop postoperative complications.

The patient is advised to carry out all of his usual activities on the first postoperative day. If he is very active, the discomfort is usually slight. Mild analgesics are usually sufficient to control any pain. If

there are any residual varices, they are injected with sclerosing solution during the postoperative visits. The elastic bandage is used from the toes to the knee for one week after removal of the sutures. If all valvular disease has been eliminated at the time of surgery and the patient is checked once a year, the leg should remain free of varicose veins. With this ambulatory regime, many patients lose only one day from work and there is considerable financial benefit for both patient and hospital.

Conclusions

As a result of recently improved ambulatory techniques, it is now possible to effectively manage practically all lower extremity venous disorders on an ambulatory basis. Superficial phlebitis can invariably be treated on an outpatient basis, while the patient carries out his normal activities. Rarely is medication or a simple high ligation of the long saphenous vein (using local anesthesia) necessary. Stasis eczema and ulceration can be effectively treated by using pressure bandages, changed once a week, on a fully ambulatory basis. Even surgery for varicose veins can be carried out, under general anesthesia, on an ambulatory basis and the patient can return to work on the first postoperative day. If bed rest is avoided, thromboembolic phenomena and other complications can be minimized. It is hoped that an ever-increasing number of medical problems in elderly patients will be managed in this ambulatory fashion.

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Management of Cancer in the Elderly

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Despite the fact that cancer incidence rises with advancing age, the management of cancer should be looked at equally in all age groups. In other words, all diagnostic and therapeutic modalities should be employed in the elderly, much as in a younger patient group.

Cancer incidence and death rates rise continuously with age and are highest among the elderly (1, 2). Even diseases with declining rates, such as gastric cancer, demonstrate increasing death rates with advancing age. Pancreatic cancer likewise exhibits an increasing death rate with advancing age.

Some diseases have their highest incidence in childhood (for example, retinoblastoma); others have bimodal patterns of distribution such as Hodgkin's disease and acute leukemia. Nonetheless, cancer incidence increases steadily with advancing age, and may be caused by the effect of a slowly acting carcinogenic agent over a long period of time. The presumed carcinogens may be chemical, physical, or biologic. Long periods of latency may be operative as in the case of asbestos workers who may develop bronchogenic carcinoma ten to twenty years after the first exposure.

Host resistance may also decline in the elderly, and might explain the increase in cancer incidence. Some disease sites predominate, such as skin cancer. If one eliminates skin cancer, then prostatic cancer accounts for one-fourth of the cancers in men, 75 years and older, while cancers of the stomach, large intestine, and rectum account for an additional 25% (3). Breast cancer in women, 75 years and older, accounts for over 20% of all cancers, while stomach, large intestine, and rectal malignancies account for a similar percentage as in men.

Symptoms of Malignant Disease in the Elderly

In the elderly, malignant disease often appears in the setting of multiple disease processes, but the physician should be alert for the disease at high risk sites. Special attention to a change in bowel habits must be given, although it is a symptom often ignored in an age group in which the alteration might be well accepted as a concomitant of advanced age. Rectal bleeding should be investigated, as should the history of alternating constipation and diarrhea.

No general agreement exists on what should constitute the annual cancer screening examination in the elderly, but most physicians agree that an annual cervical cytology smear, a monthly breast self-examination in females, and an annual rectal examination in both sexes, should be performed. The value of annual chest x-rays, proctoscopies, indeed, even annual physical examinations, is not definitively established. Specific complaints should be vigorously checked.

Older people often have difficulties with mobility and often suffer a lack of drive. Failing social competence, whether on an emotional or intellectual level, can be confused with the constitutional symptoms of an occult cancer: anorexia, weight loss, malaise, and fatigue. Toxic confusional states may supervene in malignant disease in the elderly.

A substantial number of elderly patients seek care only at the stage of advanced spread when effusions, hepatomegaly, jaundice, or abnormal masses are noted. Patient delay, caused by insufficient vigilance on the part of the family or patient, is a significant factor in cancer in the elderly.

Principles of Treatment

Lay people often look upon five-year survival rates as the criteria against which treatment regimens must be rated. But in the elderly, coexisting nonmalignant disease may influence the quality of

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life. Thus, major surgery or unpleasant side effects from chemotherapy are often contraindicated in certain instances, particularly when the patient suffers from premorbid, significant nonmalignant disease. Alternative solutions to curative surgery may be mandated if the patient is frail or debilitated. Total mastectomy may suffice for an advanced primary breast tumor if the chance of cure is remote.

Kessler recommends that total mastectomy with complete axillary dissection is indicated in patients less than eighty-years old in good health, while poor surgical risk patients should undergo total mastectomy and removal of the lower lateral axillary lymph nodes (4). Lobectomy for bronchial carcinoma has a higher mortality in the aged, and palliative measures might be preferable.

Certainly patients should be included in the decision-making process, and they should be carefully informed of the options for treatment and possible consequences of therapy. Whenever possible, patients should know what the quality of life will be like if a particular course of action is chosen.

Management of Specific Cancers

Breast cancer in women tends to be less aggressive when the disease occurs in later years, further away in time from the menopause. The disease is more aggressive in premenopausal women, especially in pregnancy. The pattern of metastasis in the elderly is also different, with a greater degree of soft tissue and bony metastases, as opposed to visceral liver and lung disease in the younger patient. Estrogen receptor activity is likewise different in postmenopausal females. A greater percentage of estrogen receptor positive primary tumors are found in the older patients, and when recurrences occur, hormonal manipulations may be more effective than chemotherapeutic intervention (5). For example, dramatic improvements in soft tissue metastasis may be seen with tamoxifen (Nolvadex®) or diethylstilbesterol in elderly patients with breast cancer patients. Hormonal therapies are often less aggressive than chemotherapeutic drugs, with fewer side effects such as white blood count and platelet suppression, stomatitis, nausea, vomiting and alopecia. But to focus for a moment on tamoxifen, hypercalcemia and a transient aggravation of existing skin lesions can occur. Reasonable assurance can be given to the elderly patient that she will be less likely to be disabled or succumb to her disease.

In men past the age of 50, the most common cancer is carcinoma of the prostate and only lung cancer deaths exceed prostatic cancer deaths in adult males. The frequency of prostatic cancer increases with advancing age. It is often not seen until autopsy, so that many cases are not appreciated clinically. The newer serum tests for clinical detection of the disease (serum radioimmunoassay for acid phosphatase) may eventually bring earlier cases to clinical attention but the true value has not been established (6). The annual rectal examination for older men continues to be the mainstay of detection when early cancer is found and, if a decision is made not to treat the disease immediately, then semianual rectal examinations, pelvic x-rays, and serum prostatic acid phosphatase determinations are necessary for follow-up. Invasive carcinomas, of course, require different approaches including prostatectomy, orchiectomy, drugs and irradiation.

Bladder cancer has the highest incidence in the older age groups. Early symptoms may be hematuria, cystitis or feelings of bladder fullness. Cystoscopy, biopsy and, occasionally, urine cytology, are important diagnostic features. New directions in the management of this cancer include combinations of drugs such as cyclophosphamide, adriamycin, and cis-platinum.

Lung cancer remains a difficult therapeutic problem in the elderly, as it does in a younger age group. The major cell type that is yielding to chemotherapeutic and radiotherapeutic intervention is oat cell cancer; but even here, median survivals beyond one year are not usually seen.

Palliative and Terminal Care

Patients with cancer, be they young or old, reach a stage of their illness when conventional therapy is ineffective. In these patients, self-dignity and self-identity are needed to fortify them in their final days. Much has been written on the hospice movement and it is beyond the boundaries of the discussion here to describe their efforts. Suffice it to say that the physician who seeks to care for aged cancer patients should have an awareness of the complex social and psychiatric as well as the biologic problems of this group. Physicians and paramedical personnel would help the patient decide where and under what circumstances death will be faced. Should the patient elect to die at home, then every effort should be made to help support the patient in his or her final hours.

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Senile Macular Degeneration: Clinical, Histopathological, and Ultrastructural Studies*

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Abstract

A review of the current status of senile macular degeneration (SMD) is presented from clinical, histopathological and ultrastructural aspects. Various stages of SMD are shown clinically and histopathologically; new ultrastructural findings of the early changes associated with the disease are also presented.

Introduction

Senile macular degeneration (SMD) is one of the leading causes of legal blindness in the United States and other countries, and is caused by partial destruction of the macular area of the retina (1). In most instances, the disease appears to be an acquired degenerative process, but in several cases, its familial nature has been described, suggesting that SMD is an autosomal dominant dystrophic disease (2). At least 25% of the elderly population, particularly persons over 70 years, are affected, and higher incidences of occurrence have been reported (3). Onset usually occurs during the sixth decade, and the median age of central vision loss is 65 years in the first affected eye, and 70 years in the second eye. Approximately 12% of the patients who have visual loss in one eye due to SMD, will develop deterioration of vision in the second eye in each succeeding year (1). SMD appears to be a caucasian disease and, while clinical and histopathological studies of large numbers of black patients in Johannesburg, South Africa revealed many instances of

retinal pigment epithelial (RPE) degeneration and drusen formation, there were no exudative or hemorrhagic forms of SMD (4). White South Africans, examined as part of the study, showed similar patterns as seen in the United States.

Anatomy of the Macular Region

Clinically, the macula is an avascular area of the retina lying 3 mm temporally and slightly inferiorly to the optic disc (5). Gross examination of the macula reveals an oval zone of yellow coloration, the macula lutea. In the center of the yellow region, an annular light reflex defines the fovea. Histologically, the macula is identified by a ganglion cell layer of more than one cell layer thick, obliquely arranged fibers of the outer plexiform layer (nerve fiber layer of Henle), a large concentration of rod-like cones among the photoreceptors, and a thick internal limiting membrane (Fig. 1). The avascularity of the central macula, or fovea, is necessary for permitting light to pass unobstructed to the photoreceptors, enabling the eye to obtain a high visual acuity.

The retinal pigment epithelium (RPE) is a single layer of cells lying beneath the sensory retina (Fig. 1). The functional importance of the RPE lies in its 1) support of the photoreceptors and provision of an absorptive surface for the light energy required by them, 2) transmission of nutrients from the choriocapillaris to the photoreceptor outer segments, 3) storage and conversion of vitamin A into

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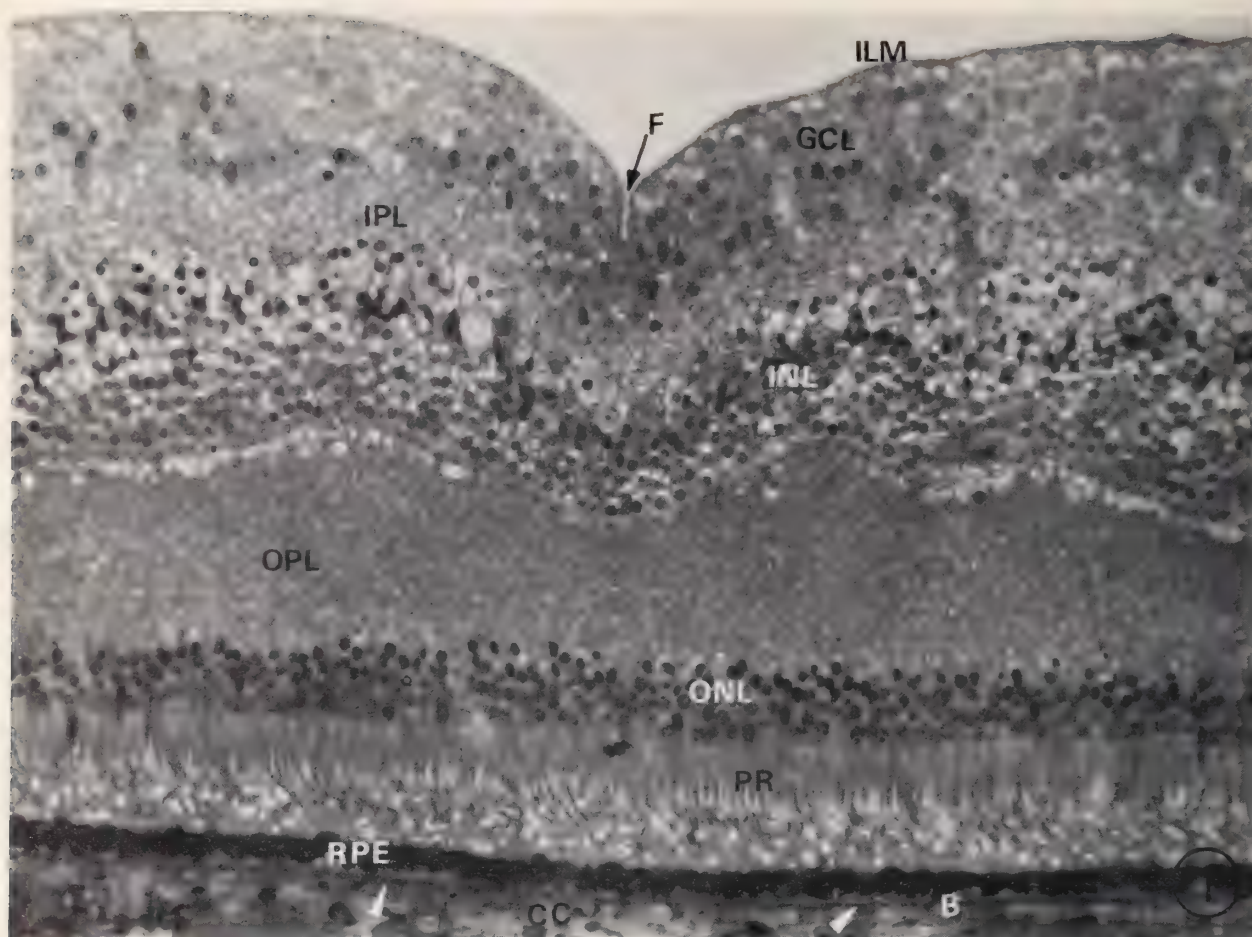


FIG. 1. Low power photomicrograph of a normal macular area. (B—Bruch's membrane, CC—choriocapillaris, F—fovea, GCL—granular cell layer, ILM—inner limiting membrane, INL—inner nuclear layer, IPL—inner plexiform layer, ONL—outer nuclear layer, OPL—outer plexiform layer, PR—photoreceptors, and RPE—retinal pigment epithelium). Toluidine blue ($\times 150$).

rhodopsin for use by the photoreceptors, 4) manufacture of extracellular mucopolysaccharides, and 5) phagocytosis and digestion of photoreceptor outer segments and other substances from the neuroretina. The RPE is separated from its vascular supply, the choriocapillaris of the choroid, by Bruch's membrane (Fig. 1), which serves as the basement membrane for the RPE and choriocapillaris and contains connective tissue.

Clinical Observations and Histopathological Correlation

The major complaint of patients with acquired macular disease is blurred vision, usually manifested as a loss of central visual acuity or central visual field. Central visual field changes, or metamorphopsia, relate to distortion in perception of size and shape of objects. The earliest detectable ophthalmoscopic changes (Figs. 2-4) in the predisiform phase of SMD⁶ are 1) loss of the foveal reflex, 2) fine stippling or mottling of the RPE in

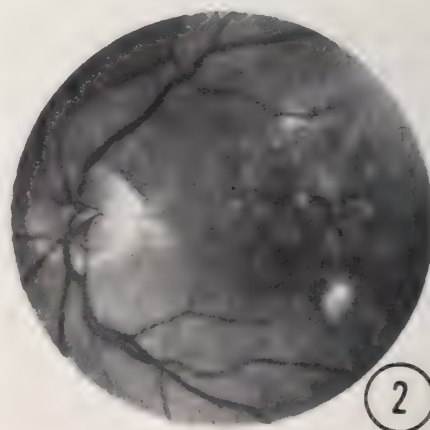


FIG. 2. Clinical photograph of left eye showing pigment mottling and drusen formation in the macula.

the macular area, and 3) development of central drusen. Drusen, defined histochemically as deposits of mucopolysaccharides and lipids between RPE and Bruch's membrane, appear ophthalmoscopy-



FIG. 3. Late phase of a fluorescein angiogram (59.1 seconds), right eye showing "window defects" (arrow) in the RPE.



FIG. 4. A fluorescein angiogram of the left eye reveals numerous drusen in the posterior pole.

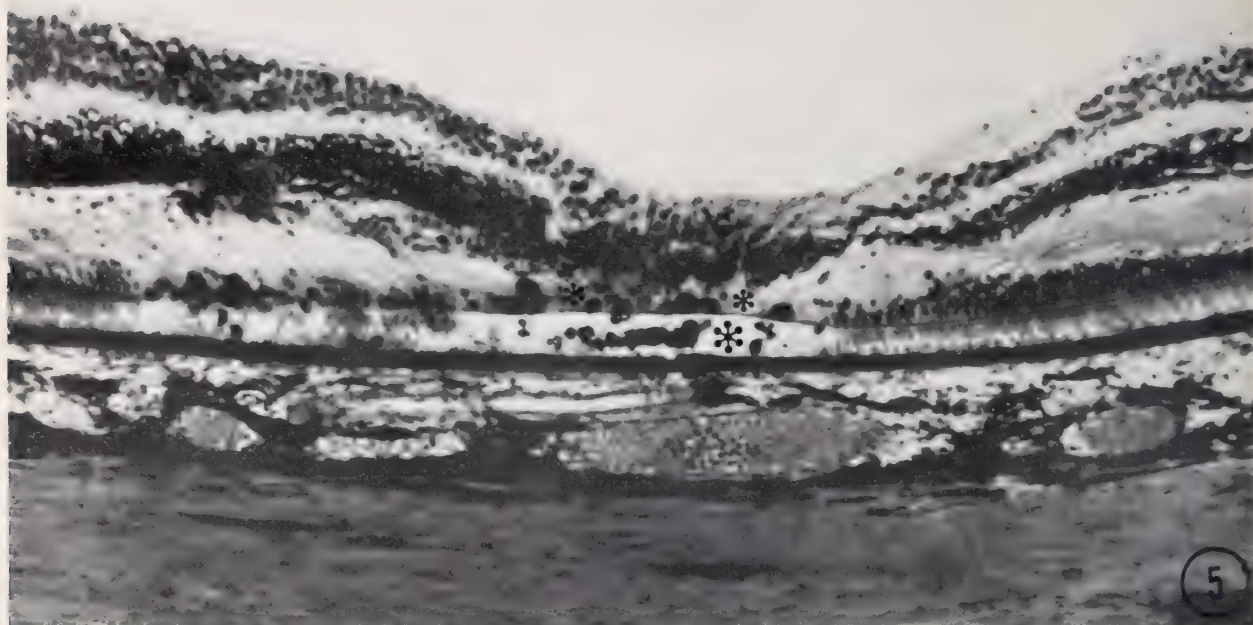


FIG. 5. Low power photomicrograph showing RPE degeneration with loss of overlying photoreceptors (large*) and outer nuclear layer (small*). Hematoxylin-eosin ($\times 200$).



FIG. 6. Higher power micrograph of atrophic retina with central loss of RPE (*) and photoreceptor degeneration (arrow). Toluidine blue ($\times 550$).

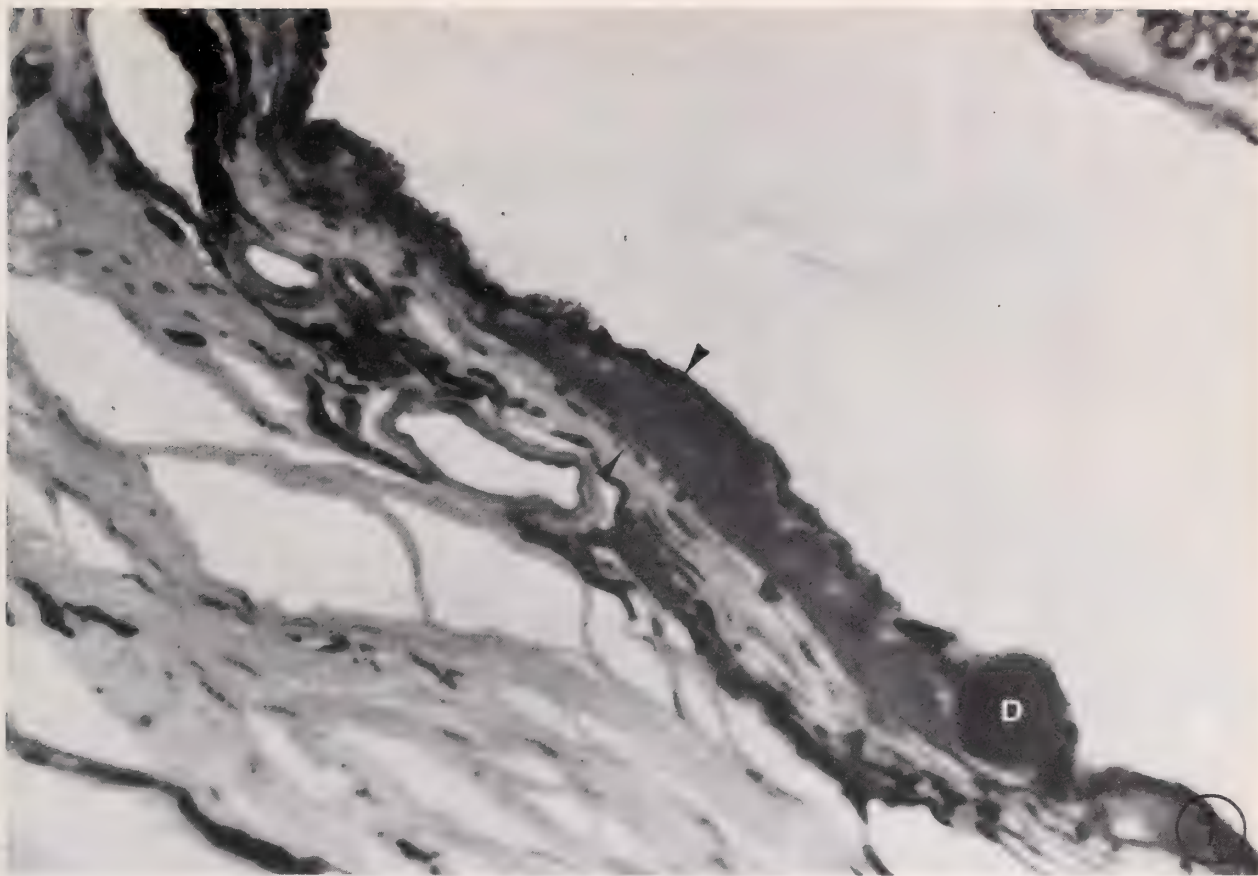


FIG. 7. An area of diffusely thickened (between arrows) Bruch's membrane with drusen (D) formation. The retina shows artifactual separation.

Periodic acid-Schiff ($\times 400$).

cally as yellow-white, discrete round, often multiple lesions. Their color is dependent upon the extent of thinning of the overlying RPE. Drusen enlarge with time and may eventually calcify in which case they present a glinting or glistening appearance (7). Histopathologic studies of eyes at the predisciform stage of SMD (Figs. 5-8) reveal 1) irregular thickening and calcification of Bruch's membrane, 2) thickening and hyalinization of the intercapillary columns of the choriocapillaris (basal laminar deposits), 3) narrowing of choriocapillary lumen (8), 4) diminution of cellularity (8) of the capillaria, 5) deposition of granular or homogeneous material (drusen) under the RPE, and 6) minimal photoreceptor degeneration.

As SMD evolves, ophthalmologic examination reveals the presence of serous detachments of the RPE (Figs. 9-11) in association with 1) atrophy of the RPE and 2) multiple drusen. Serous fluid may pass into the subretinal space and produce a serous detachment of the sensory retina (Figs. 12-14). In time, fine new blood vessels may invade the subpig-

ment epithelial space (Figs. 15-18) (9). Bleeding, confined to the subpigment epithelial space results in a dark grey mound in the macular area (Fig. 19). The blood may, however, extend into the vitreous cavity. Bleeding into the subretinal space from the subpigment epithelial space produces a red halo about the central dark lesion (Fig. 20). The changes of SMD associated with the hemorrhagic stage are a result of the subretinal pigment epithelial neovascularization (9). The new vessels formed are presumably derived from choroidal vessels that enter the subretinal pigment epithelial space through breaks in Bruch's membrane. They may be visualized ophthalmoscopically through defects in the atrophic RPE or, more clearly, during fluorescein angiography, as a fine network of blood vessels. As the condition progresses, new vessels invade the subretinal space (Fig. 21). Subretinal hemorrhage and fatty exudates are often signs of subretinal neovascularization. With even further progression of the condition, the neovascular membranes tend to leak and bleed, leading to serous and hemor-

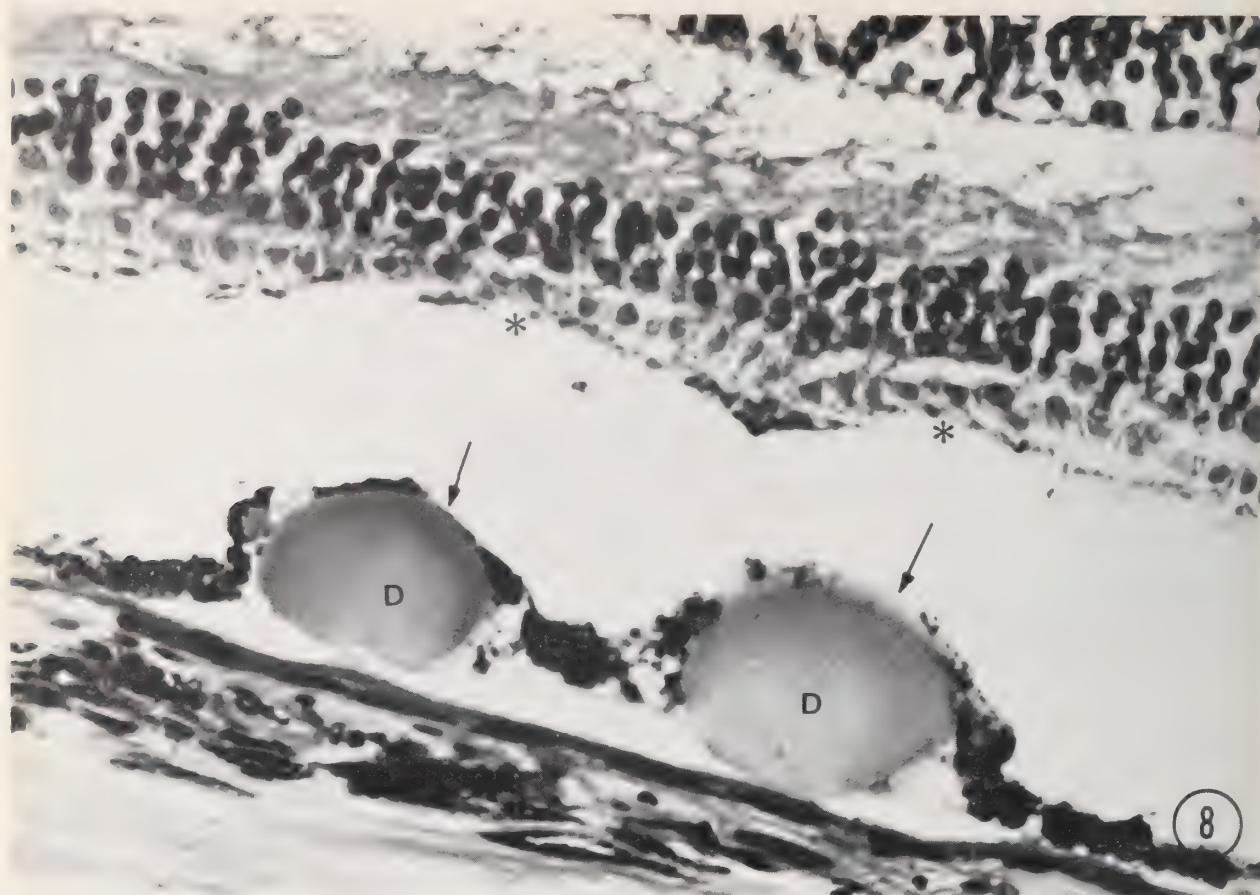


FIG. 8. Two focal drusen (D) are covered by an attenuated RPE (arrow). Note thinning of photoreceptors overlying the drusen (*). Hematoxylin-eosin ($\times 500$).

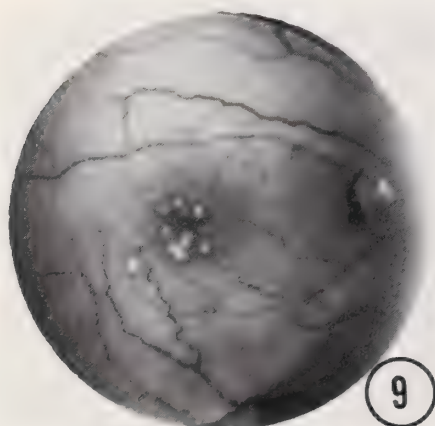


FIG. 9. Several focal serous detachments of the RPE, left eye.



FIG. 10. A photograph of the right eye showing serous detachment of retina just superior to the macula. A row of intraretinal exudate is present.

rhagic detachment of the RPE. The hemorrhage may resorb, or dissect under or into both the retina (Figs. 22, 23) and overlying vitreous cavity.

The presence of blood under the RPE or retina stimulates the proliferation of fibrous connective tissue and hyperplasia of RPE cells (Fig. 24). The

end result of this process is a white or pigmented subretinal scar (Figs. 25-28) with cystoid degeneration of the macular area and loss of the photoreceptors. Alternatively, during the evolution of this process, profuse exudation from vessels may occur;

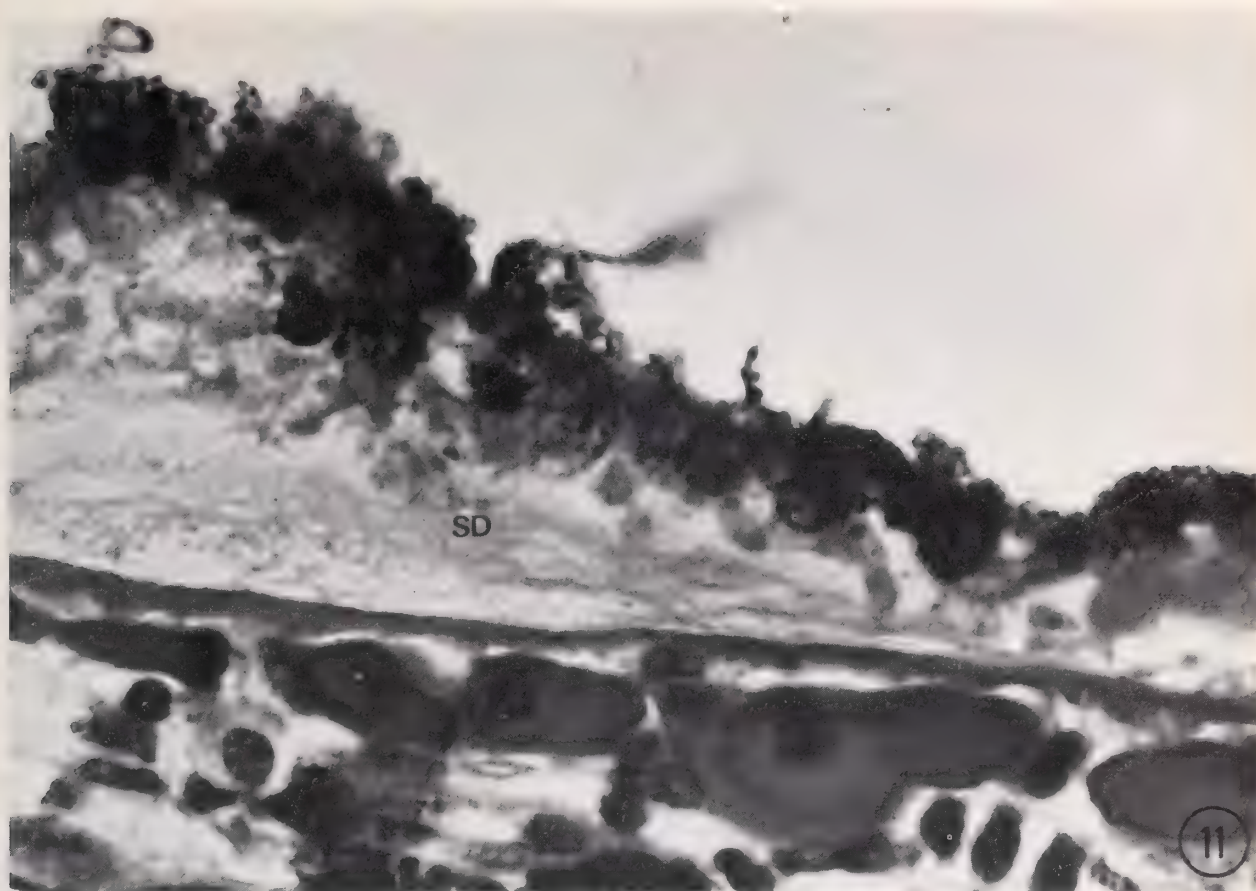


FIG. 11. An area of serous detachment (SD) of RPE. Hematoxylin-eosin ($\times 1000$).

this leads to massive subretinal and intraretinal serous exudates often containing large amounts of lipid (Fig. 29). Clinically, at this final stage of SMD, there is a significant loss of central vision, often to levels of legal blindness.

Electron Microscopic Observations

The comprehensive ultrastructural anatomy of the cell types and their relationships to each other in SMD eyes has only been touched upon, but fine structural changes in retina and choroid as a function of age are well-documented (5, 11–15). Electron microscopy of SMD eyes has so far shown the basal linear deposits to contain banded fibrous or spindle-shaped residues, residing in a granular matrix, and found between the basement membrane and cell membrane of the RPE (16). Small amounts of basal linear deposits are considered to be normal in the aging eye (16).

Most of the age-related changes associated with normal eyes appear to be present in SMD eyes also (10, 13, 16). Fine structural changes in Bruch's



FIG. 12. Clinical photograph of the right eye revealing intraretinal hemorrhage (arrow) and serous detachment of the retina (*).

membrane, as a function of age, are marked by a progressive accumulation of vesicles; an increase in dense collagen fibers, normal, and wide-spacing collagen; banded fibers; calcification; increased density

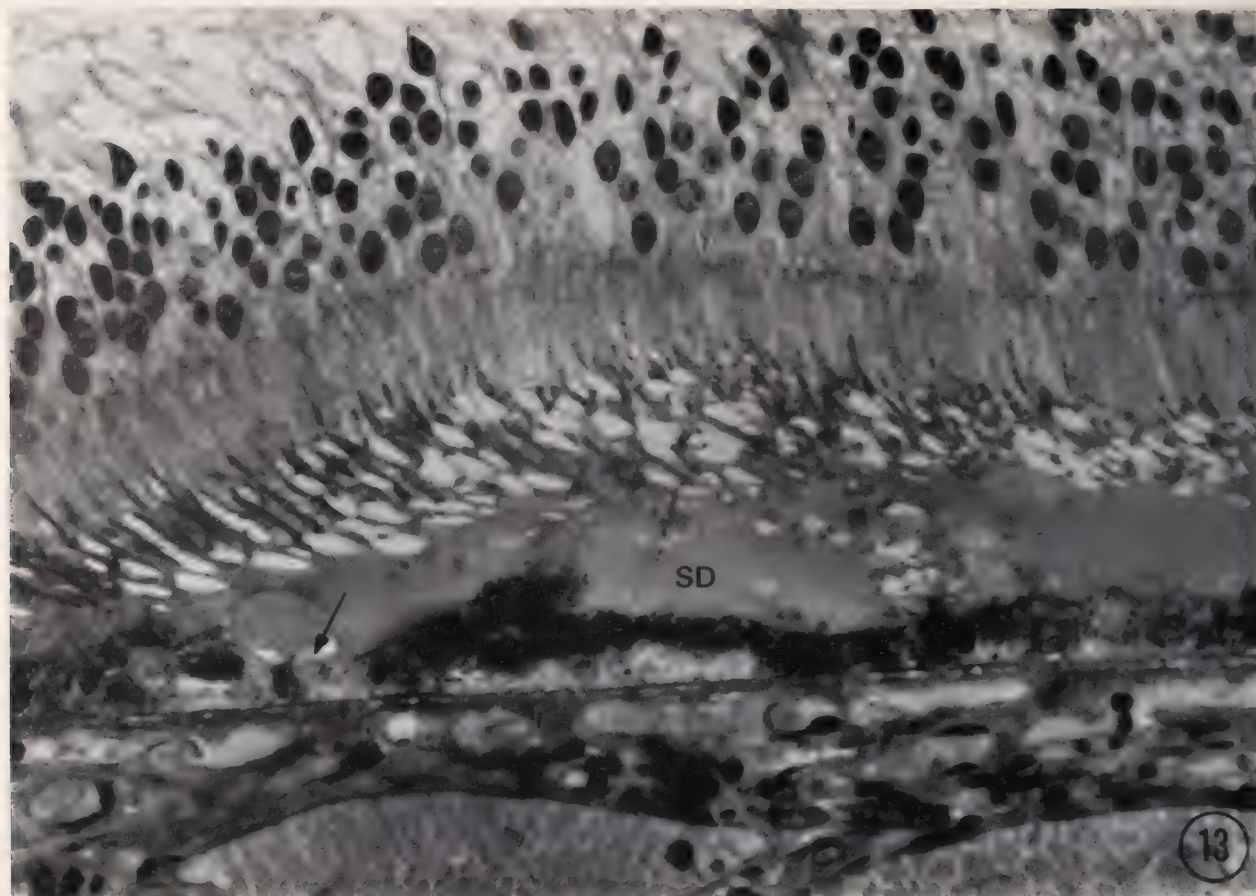


FIG. 13. An area of RPE (arrow) damage with serous detachment (SD) of overlying retina. Hematoxylin-eosin ($\times 550$).



FIG. 14. The retina shows serous detachment (SD) in the macular area with cystoid macular edema (*). Hematoxylin-eosin ($\times 150$).



FIG. 15. Arteriovenous phase of angiogram, right eye, revealing subretinal neovascularization (*), subretinal hemorrhage (white arrow) and serous detachment of the retina (large *).



FIG. 16. Light micrograph showing calcification (c) and fragmentation of Bruch's membrane (arrow). Note area of sub-RPE neovascularization at right (SNV). Hematoxylin-eosin ($\times 800$).

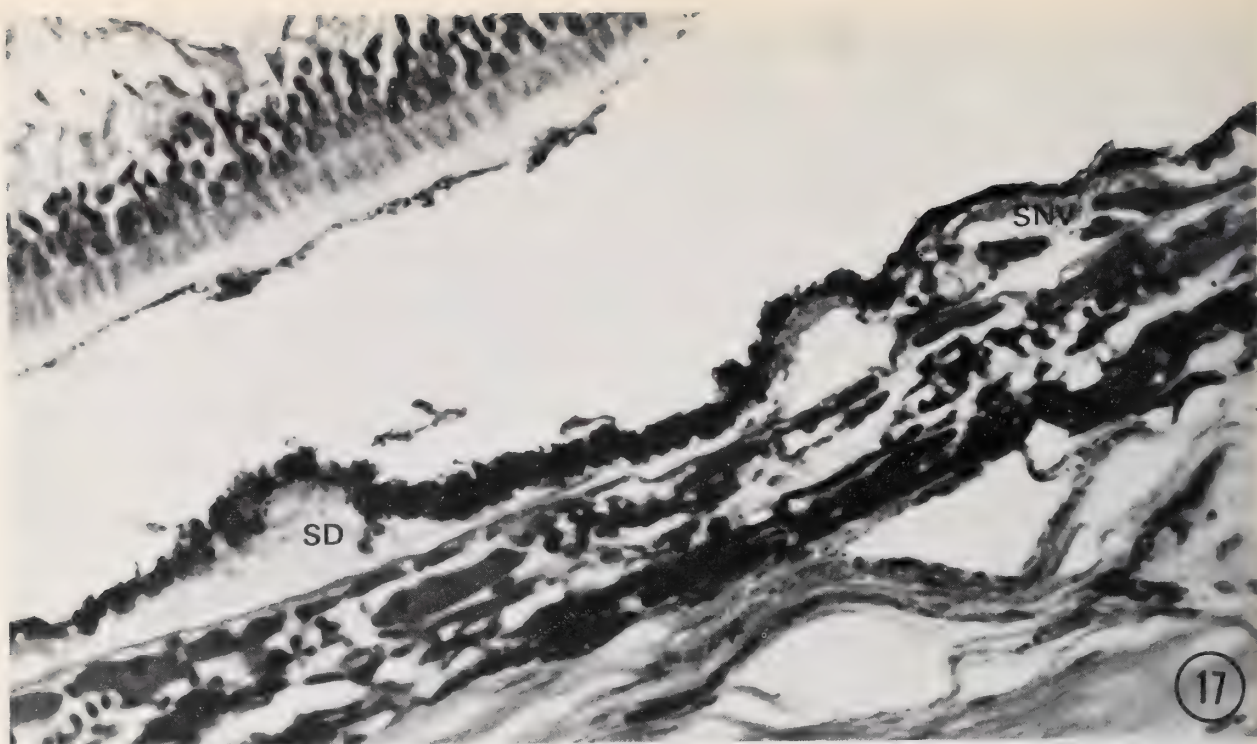


FIG. 17. Low power micrograph of serous detachment (SD) of RPE at left, and sub-RPE neovascularization (SNV) at right. Hematoxylin-eosin ($\times 250$).

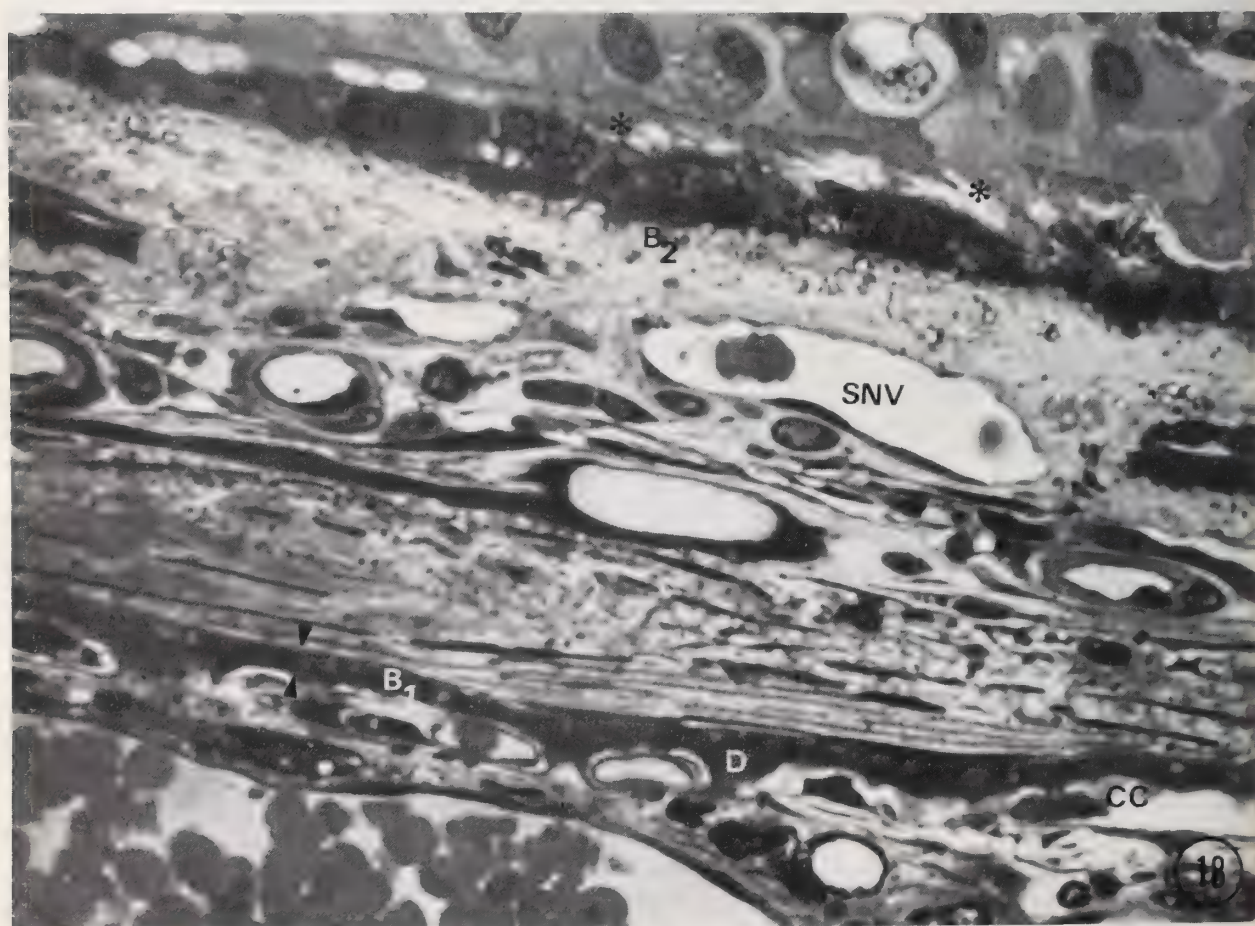


FIG. 18. A higher power photomicrograph showing an area of subretinal pigment epithelial neovascularization (SNV) between Bruch's membrane of choriocapillaris (B_1) and the RPE (B_2). Note degenerating outer segments (*), and thickening of Bruch's membrane (arrows) especially the deposits (D) between choriocapillary vessels (CC). Toluidine blue ($\times 850$).



FIG. 19. Late phase fluorescein study shows hemorrhage (black arrow) in retina with subretinal exudation and striae (white arrow). Disregard dark line.

in the core of elastic fibers; accumulations of tube-like structures; and accumulation of drusen (5, 12, 13). Normal aging RPE cells are characterized by increased lipofuscin, vesicles, granular material, fibrous formations between the basal infoldings, and alteration of basal infoldings (5, 12, 13).

The choriocapillaris in SMD showed little alteration from its normal fine structure. Profiles of endothelial cells showed well-developed fenestra, with no increase in the thickness of the basement membranes, or changes in the cell cytoplasm (Fig. 30). Debris, associated with the outer collagenous zone of Bruch's membrane, was seen extending between capillaries (Fig. 31). Ultrastructurally, this debris, responsible for the increase in thickness of Bruch's membrane as seen by light microscopy, was characterized by vesicles, tube-like structures, banded structures, and membrane-bound dense bodies (Fig. 32, 33). Occasional drusen, composed of material similar to the debris described above (Fig. 34), were seen extending from the inner aspect of Bruch's membrane, and displaced the RPE inwardly. RPE showed a reduction in basal infoldings, with banded structures occasionally seen between those few basal infoldings observed (Fig. 32, 33). Melanin was found predominantly in the basal re-

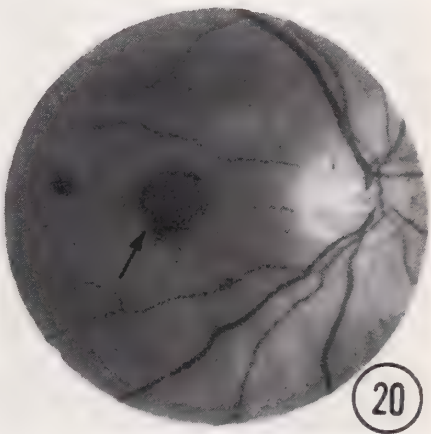


FIG. 20. A hemorrhagic detachment of the RPE is surrounded by a ring of intraretinal hemorrhage (arrow).

gion of the cell, and few profiles of rough or smooth endoplasmic reticulum were observed. There appeared to be no interaction of outer segments and RPE, and only a few apical cell processes were observed (Fig. 31, 35). Morphologically, there was some evidence of phagocytosis of debris seen adjacent to the apical surface of the RPE (Fig. 31) and outer segments showed occasional swelling and vesiculation and corrugation of the discs (Figs. 31-36).

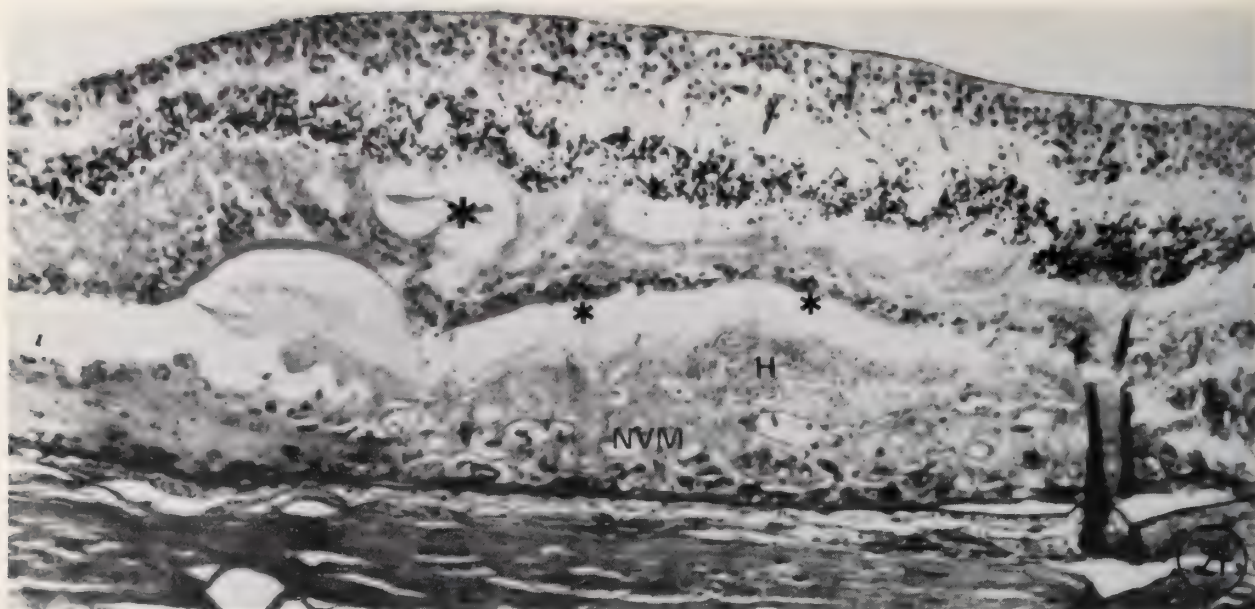


FIG. 21. A large subretinal neovascular membrane (NVM) with hemorrhage (H) is present underneath a partially atrophic retina. Note loss of photoreceptors centrally (small *) and cystoid macular edema (large *). Luxol fast blue ($\times 250$).

Conclusions

It is still not known why the macular region of the retina is predisposed to the development of SMD. Aging, race, and perhaps, heredity are important factors, but others, such as vascular supply, long term effects of light and mechanical trauma cannot be discounted. The diagrammatic summary of the natural history of SMD (Fig. 37) indicates serious consequences of SMD in relation to the development of serous detachment of the RPE. Perhaps, as a result of further separation of the outer retina from its blood supply (the choriocapillaris), new vessels enter the subpigment epithelial space and eventually lead to the development of serous and hemorrhagic detachments and their sequelae. This occurs in association with aging changes in Bruch's membrane. It is early recognition of the subpigment epithelial neovascular net, through use of fluorescein angiography, that may have significant salutary benefits to the patient with SMD. The obliteration of these nets, using laser photocoagulation, may, in some cases arrest the progression of the condition (17).

Acknowledgments

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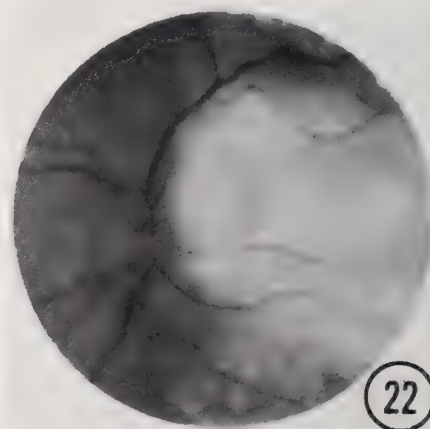


FIG. 22. Clinical photograph of left eye showing massive subretinal exudation covering the entire posterior pole.

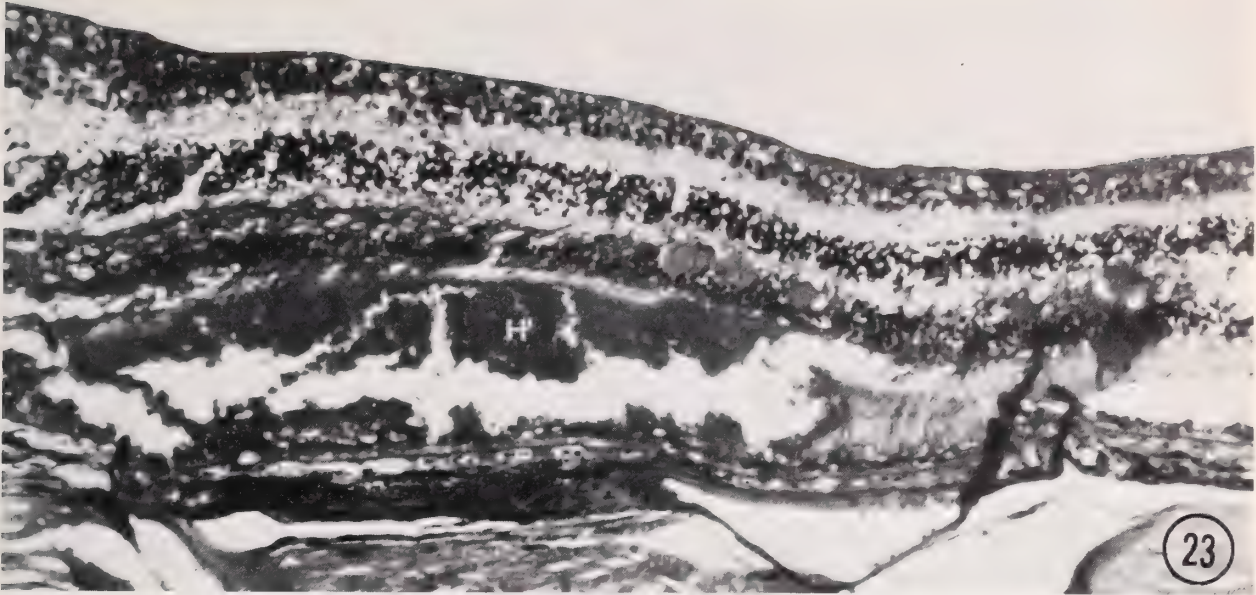


FIG. 23. A hemorrhagic detachment of the macula (H).
Hematoxylin-eosin ($\times 250$).

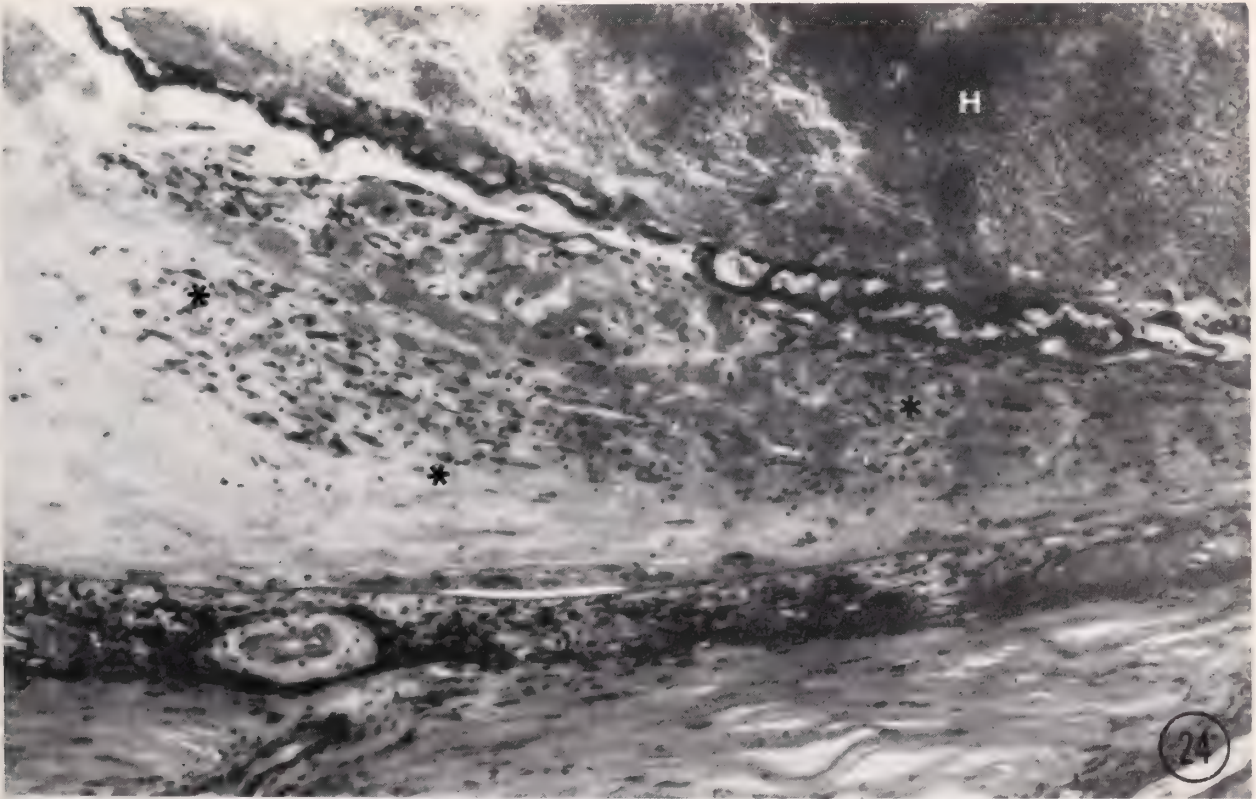


FIG. 24. Early organization (*) of subretinal pigment epithelial hemorrhagic detachment, and hemorrhagic detachment of (H) retina
above and to the right.
Hematoxylin-eosin ($\times 500$).



FIG. 25. Photograph of left eye containing massive intraretinal hemorrhage (large arrow) and exudates (small arrow).



FIG. 26. Massive subretinal fibrovascular membrane (*). Note folding of inner retina. Luxol fast blue ($\times 150$).



FIG. 27. Higher power micrograph of subretinal fibrovascular membrane. Note areas of RPE hyperplasia (arrow), and atrophy of the outer retina (*). Toluidine blue ($\times 250$).

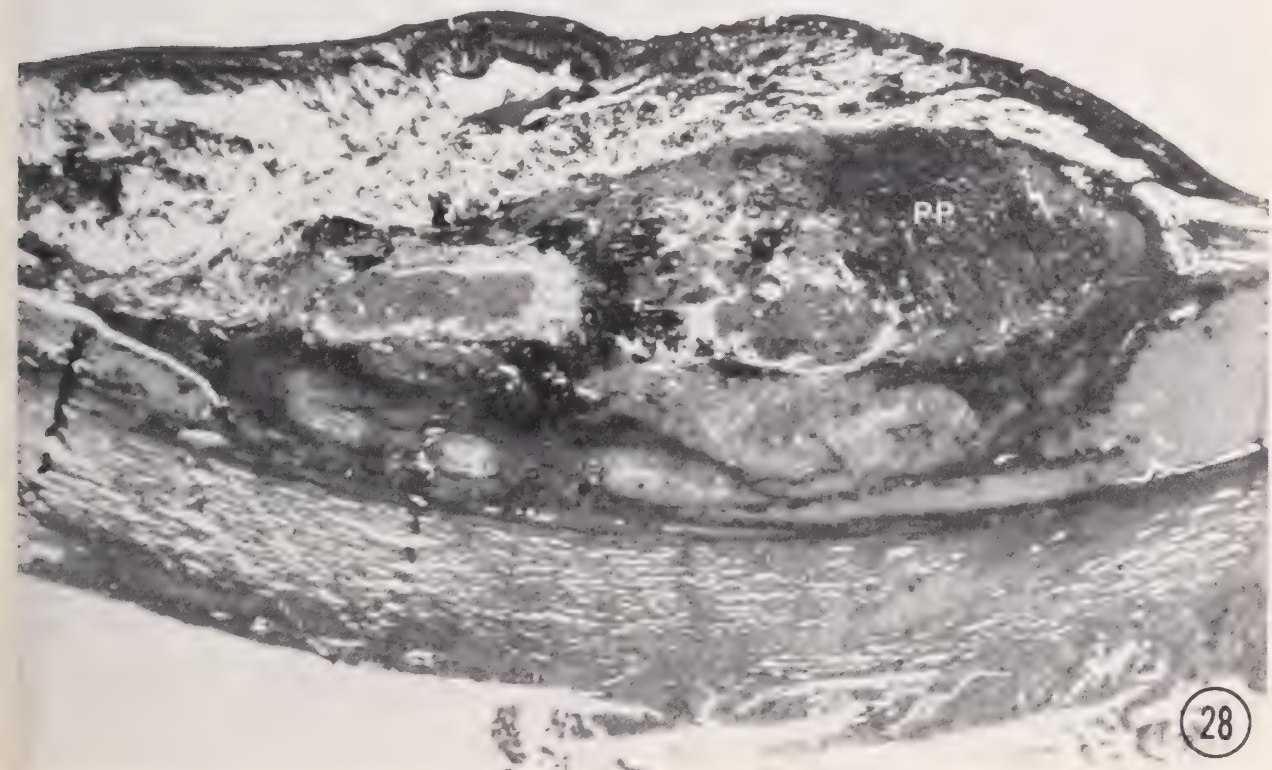


FIG. 28. A large pigmented subretinal fibrous plaque (PP). Pigmentation due to deposition of hemosiderin, RPE hyperplasia and free melanin granules. Hematoxylin-eosin ($\times 100$).

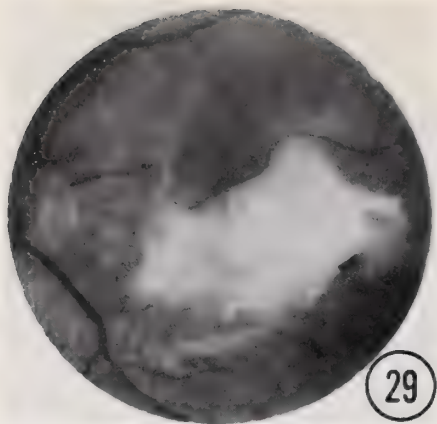


FIG. 29. Clinical photograph of left eye showing a fibrous subretinal scar.

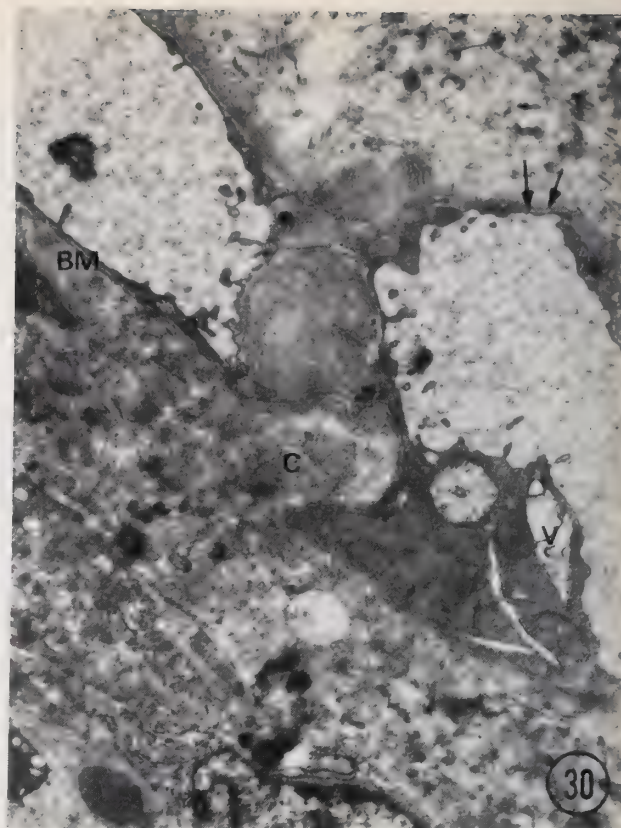


FIG. 30. Electron micrograph of portion of the choriocapillaris in SMD eye showing an uncharacteristically irregular appearance of the capillary lumen. Vacuolization (V) of the endothelium can be seen along with a typically fenestrated wall (arrows). Basement membrane (BM) appears characteristic. Masses of collagen (C) fibrils can also be discerned. Uranyl-lead ($\times 9900$).

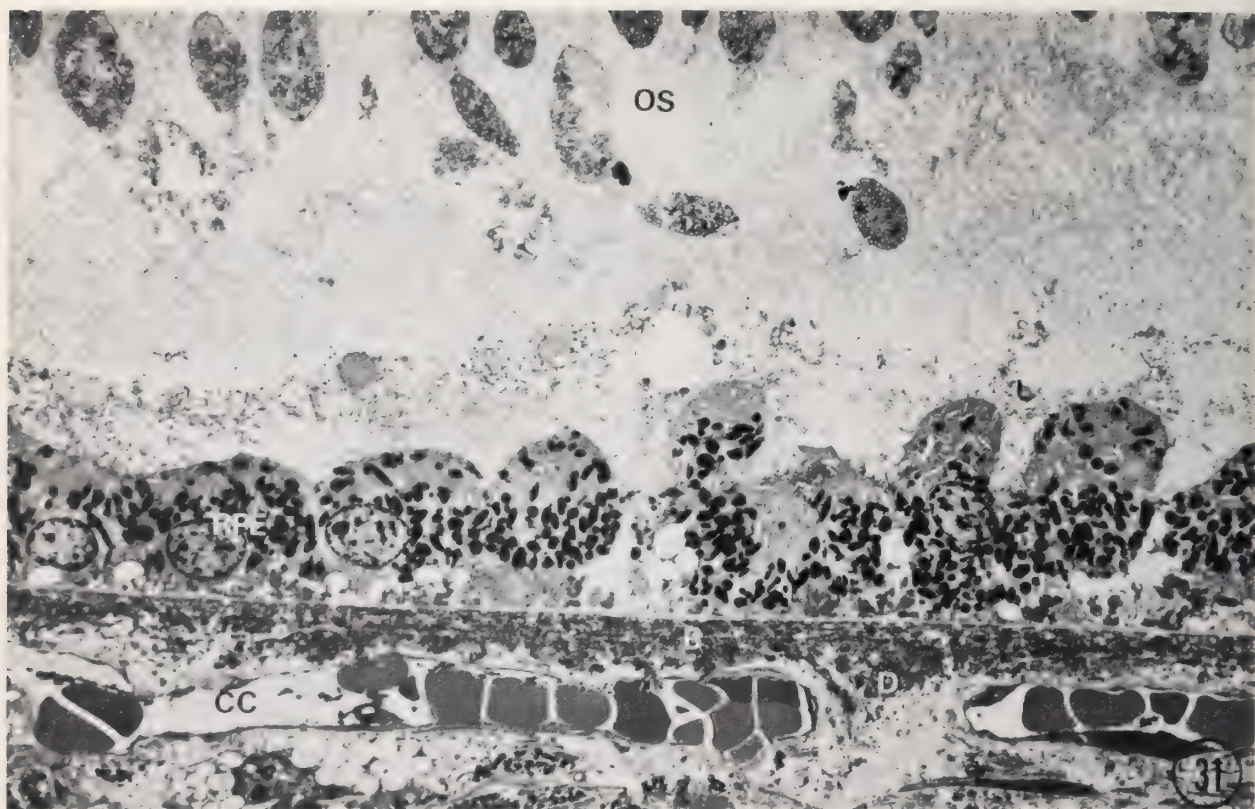


FIG. 31. Low magnification electron micrograph of choroid, Bruch's membrane (B), retinal pigment epithelium (RPE), and outer segments (OS) of the macular area in the eye of a patient with SMD. Note lack of interaction of outer segments, debris (D) in Bruch's membrane, and normal appearance of choriocapillaris (CC). Uranyl-lead ($\times 1400$).

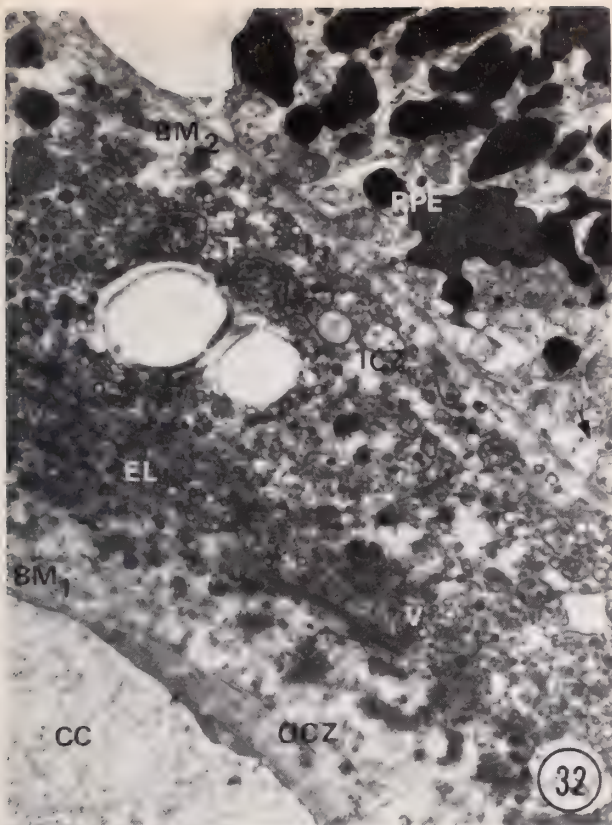


FIG. 32. Electron micrograph of choriocapillaris (CC), Bruch's membrane (BM₁-basement membrane of choriocapillaris, BM₂-basement membrane of RPE, EL-elastic layer, ICZ-inner collagen zone, OCZ-outer collagen zone), and basal region of retinal pigment epithelium (RPE). Numerous vesicles (V) and tube-like structures (T) are seen, with the RPE cell showing only occasional basal infoldings (arrow). Uranyl-lead ($\times 9600$).

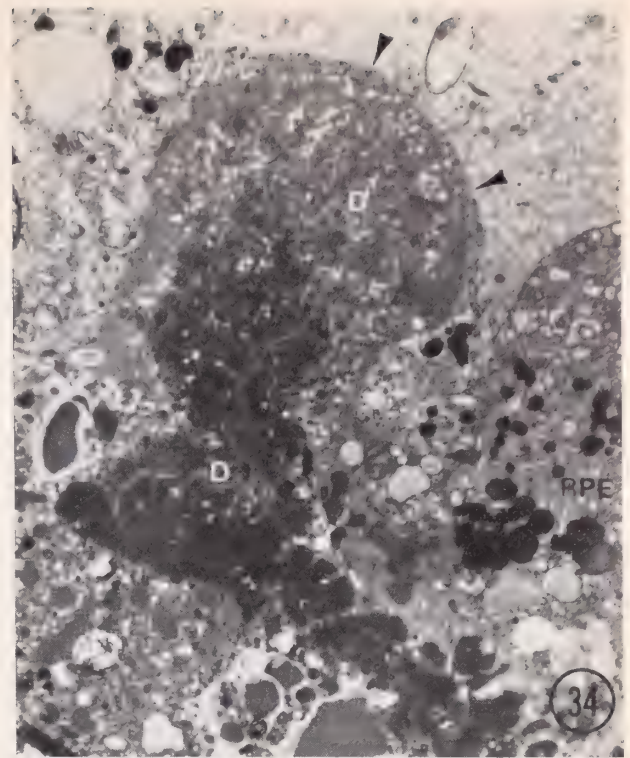
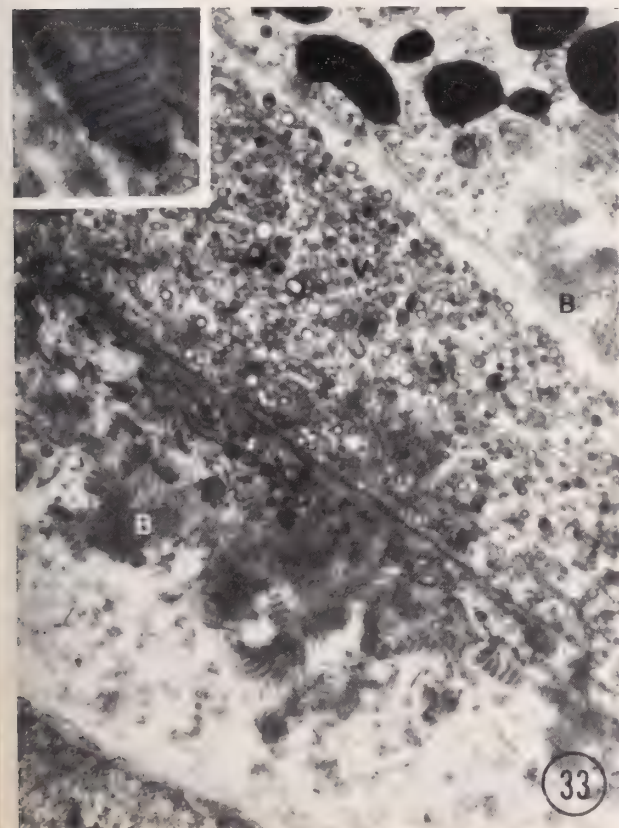


FIG. 34. Electron micrograph of RPE and Bruch's membrane which shows drusen (D) extending into and displacing the RPE (arrow) toward the detached outer segments. Uranyl-lead ($\times 3900$).

FIG. 33. Electron micrograph of banded structures (B) occasionally seen in Bruch's membrane, which can also be seen within the basal infoldings of RPE cells. Note numerous vesicles (V) in the inner collagenous zone of Bruch's membrane. Uranyl-lead ($\times 12000$). (inset)

Higher magnification of banded structures. Uranyl-lead ($\times 18000$).

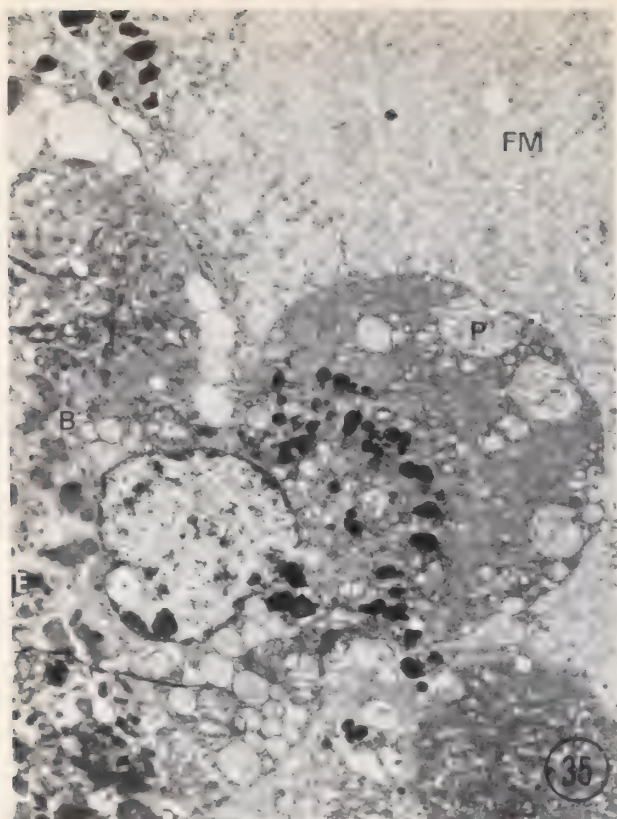
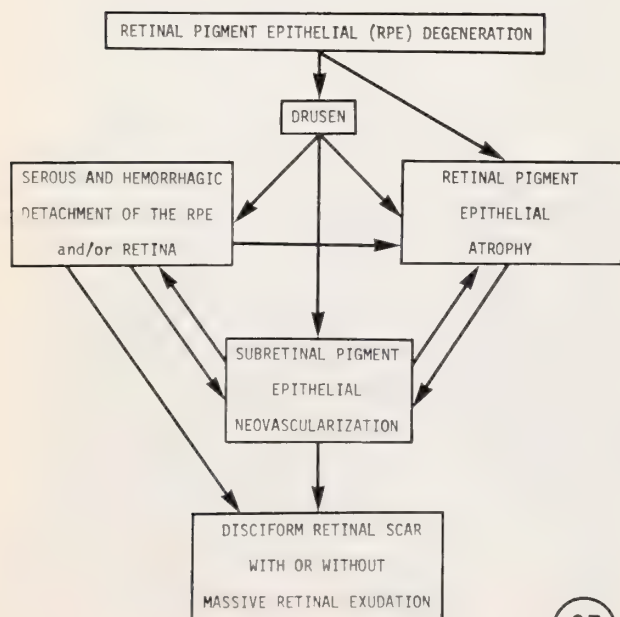


FIG. 35. Electron micrograph of SMD eye showing disrupted organization of Bruch's membrane in apposition to RPE cell. The RPE cell shows none of the typical processes nor is it associated with any outer segments. Here, within the normally more organized configuration of Bruch's membrane, numerous vesicles (V), elastic material (E), elongated banded structures (B), and other debris and atypical structures abound. Evidence of phagocytosis (P) of flocculent material (FM) can be seen. Uranyl-lead ($\times 3500$).



FIG. 36. Electron micrograph of inner (IS) and outer (OS) segments of cones in the macular area of a patient with SMD. The outer segment discs show marked corrugation (C) and vesiculation (V). Swollen mitochondria (M) are seen in the inner segments. A flocculent material (FM) can be seen in the extracellular space between photoreceptors. Uranyl-lead ($\times 9200$).

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FIG. 37. Proposed flow diagram for SMD. (Modified from Green and Key (10).

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ERRATUM

In "The On-Off Phenomena in Parkinson's Disease" by Larry Goldstein, B.S. (Vol. 47, Issue 1, January-February 1980, p. 82), *Results* section: The level of significance was inadvertently misprinted. In paragraph two, sentences 2 and 4, the level of significance should read $p < .01$.

Effect of a Single Long-acting Testosterone Injection on Growth Hormone Responses in Delayed Puberty

HARVEY SUDZIN, M.D. AND FREDDA GINSBERG-FELLNER, M.D.

Abstract

The effect of testosterone on growth hormone responses to insulin-induced hypoglycemia and L-DOPA was studied in a 16½-year-old male with small stature and delayed puberty. Augmentation of growth hormone responses to both stimuli was demonstrated after a single low-dose injection of long-acting testosterone. This case substantiates the usefulness of low-dose testosterone priming in determining growth hormone reserve, and documents for the first time the reversibility of the testosterone effect.

Introduction

In boys with small stature, the differentiation between delayed puberty and growth hormone deficiency often is difficult on the basis of clinical observation alone. Laboratory tests are helpful but must be interpreted with caution. The results of growth hormone stimulation tests often are equivocal (1-5). It, therefore, has been suggested that, for diagnostic purposes, a short course of testosterone may be helpful by augmenting the growth hormone responses to insulin-induced hypoglycemia and arginine infusion (3-6). A case is presented in which a single low-dose injection of long-acting testosterone augmented the growth hormone responses to both insulin-induced hypoglycemia and oral L-DOPA. The reversibility of these effects was also demonstrated.

Case Report

E.C., a 16½-year-old Ecuadorian male, was referred to The Mount Sinai Hospital for further evaluation of small stature, delayed puberty, and

possible growth hormone deficiency. The diagnosis of delayed puberty had been made on the basis of physical examination and a delayed bone-age (7) of 12 years. An inadequate growth hormone response had been obtained after insulin-induced hypoglycemia (maximum growth hormone level of 2.2 ng/ml after a >50% fall in serum glucose using 0.1 units/kg of regular insulin intravenously). Other laboratory data had included a normal complete blood count, urinalysis, erythrocyte sedimentation rate, serum sodium, potassium, chloride, calcium, phosphorous, and blood urea nitrogen (BUN), T₃, T₄, TSH, skull x-rays, tomograms of the sella turcica, and visual fields.

On physical examination, the patient had a height of 149.9 centimeters (<3rd percentile) and weighed 50.0 kilograms (5th percentile). Old growth records were not available, but according to the patient's parents, there had been no appreciable growth since the age of 12. There was absence of pubic, axillary, and facial hair. Testicular volume was 3 cubic centimeters by orchidometer (8) (Prader). Penile length was 6 centimeters. It was of note that the patient's older brother allegedly had not developed pubic hair until the age of 18 years, and at age 20 years was a fully developed adult male with a height of 162.6 centimeters. Maternal and paternal heights were 159.9 centimeters and 165.1 centimeters, respectively.

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Materials and Methods

After obtaining informed consent from the patient and his parents, growth hormone stimulation tests using insulin and L-DOPA were performed in the same manner on three different occasions: 1) at initial evaluation, 2) two weeks after a single injection of testosterone enanthate 100 milligrams intramuscularly, and 3) seven weeks after the single injection of testosterone. All testing was done in the basal state, after an overnight fast, with the patient in the recumbent position. An intravenous line for blood sampling was kept patent with a slow drip of 0.9% isotonic saline through a 21-gauge butterfly needle. Regular insulin (0.1 units/kilogram) was given intravenously at time 0 and L-DOPA (500 milligram crushed tablet) was given orally at 60 minutes. Blood samples for glucose, growth hormone, and cortisol were obtained at 0, 15, 30, 45, 60, 75, 90, 105, 120, 150, and 180 minutes. Growth hormone (9) and cortisol were measured by radioimmunoassay techniques. At time 0, plasma testosterone, FSH, and LH were measured also by radioimmunoassay.

Results

The results of all blood tests are shown in Table I and the growth hormone responses are illustrated in Figure 1. In all instances, the fall in plasma glucose after administration of insulin was >50%. At initial evaluation and seven weeks after the single injection of testosterone, the plasma testosterone levels were 55.0 ng/dl and 78.8 ng/dl, respectively. At these low levels, the growth hormone responses both to insulin-induced hypoglycemia

and L-DOPA were similarly blunted; in neither instance did the plasma growth hormone rise over 5.9 ng/ml (normal values in our laboratory in children and adolescents are greater than 7 ng/ml). In contrast, two weeks after the injection of testosterone, the plasma testosterone was 278.5 ng/dl, and the growth hormone responses both to insulin and L-DOPA were normal; maximum growth hormone after insulin was 16.2 ng/ml, and after L-DOPA, 14.2 ng/ml. Serum FSH and LH were in the prepubescent range on all three occasions.

Discussion

Boys with small stature and delayed puberty frequently are suspected of having growth hormone deficiency. Stimulation of growth hormone in these boys with insulin and arginine has been shown often to give equivocal results, making it difficult to exclude growth hormone deficiency (1-5). In order to better determine growth hormone reserve in these patients, it has been suggested that pretreatment with androgens, followed by repetition of stimulation tests, be performed. The basis for this has been the demonstration of augmentation of growth hormone responses both to insulin-induced hypoglycemia and arginine infusion after pretreatment both with high and repetitive doses of oral and intramuscular testosterone preparations (1-4). In fact, Deller et al (1) reported one case in which the augmented growth hormone response to hypoglycemia was demonstrable months after androgen administration was discontinued. Whether or not this prolonged enhancement was coincidental with the induction of spontaneous pubertal development

TABLE I
Summary of Laboratory Data

	Initial Evaluation			Two Weeks after Testosterone Enanthate 100 mg Intramuscularly			Seven Weeks after Testosterone		
Plasma testosterone (ng/dl)	55.0			278.5			78.8		
Serum FSH (mIU/ml)	1.1			1.0			0.4		
Serum LH (mIU/ml)	0.6			0.6			0.3		
Time (minutes)	Glucose (mg/dl)	GH (ng/ml)	Cortisol (μg/dl)	Glucose (mg/dl)	GH (ng/ml)	Cortisol (μg/dl)	Glucose (mg/dl)	GH (ng/ml)	Cortisol (μg/dl)
0	97	2.3	23.5	83	3.1	14.5	104	0.8	20.0
15	58	4.9	22.0	61	16.2	14.5	47	0.8	16.5
30	38	5.0	18.0	36	9.6	12.5	35	0.8	14.0
45	69	5.9	19.0	50	8.3	16.0	52	4.5	24.5
60	76	3.2	12.0	66	8.4	16.0	75	4.8	26.5
75	82	2.7	13.0	80	5.9	15.0	93	3.7	30.0
90	96	1.8	10.0	89	9.2	15.5	93	5.0	21.0
105	119	1.8	22.5	84	14.2	15.5	92	4.7	18.5
120	122	4.4	26.5	94	9.8	18.0	95	2.2	18.0
150	116	4.2	27.5	97	6.6	21.5	105	0.8	12.0
180	96	3.0	28.0	99	3.3	15.0	99	0.8	9.0

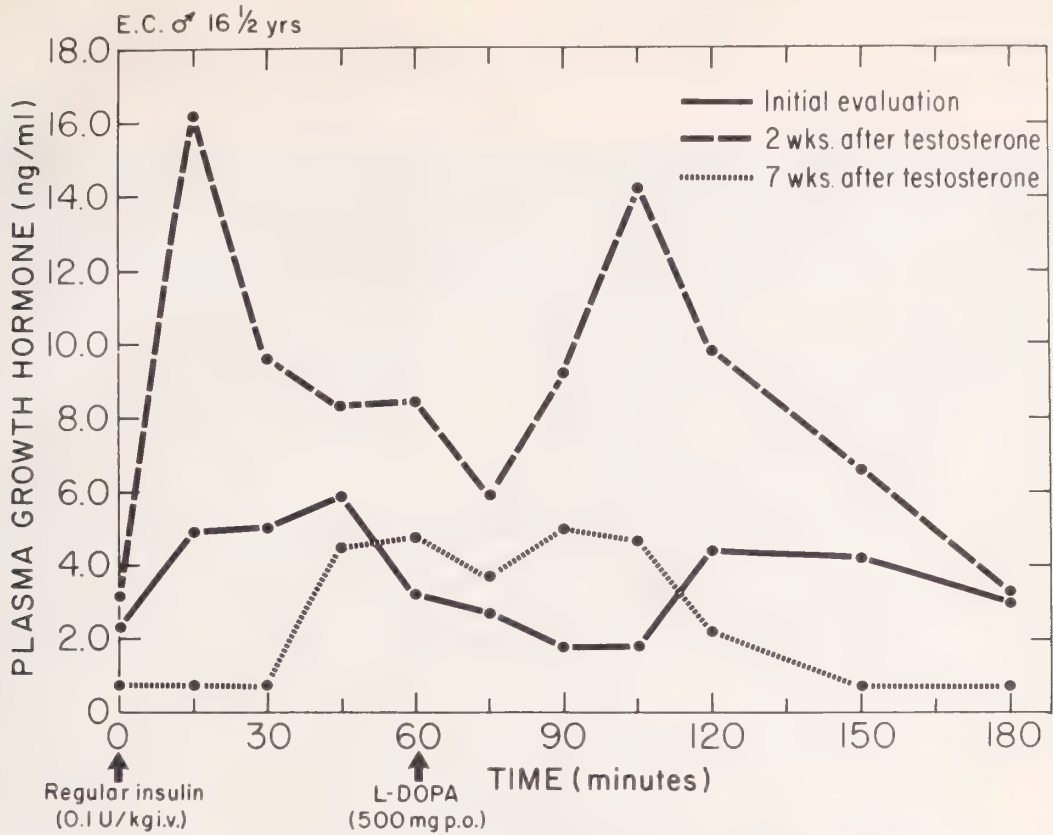


FIG. 1. Growth hormone responses to insulin-induced hypoglycemia and L-DOPA

is speculative, as measurements of FSH, LH, and plasma testosterone were not reported. That the stage of puberty may indeed be important in assessing growth hormone reserve in boys with delayed adolescence is also substantiated by Penny and Blizzard (5) who showed normal growth hormone responses to hypoglycemia and arginine infusion in three males ages 14.3 to 19 years only after they had reached Tanner Stage IV sexual development.

The data from the above patient have several implications regarding growth hormone responses in boys with small stature and delayed puberty. First, in addition to subnormal responses to other stimuli, the growth hormone response to L-DOPA also may be blunted, and may be augmented by pretreatment with testosterone if growth hormone reserve is adequate. Second, the effect of testosterone on growth hormone responses both to insulin-induced hypoglycemia and L-DOPA appears to be reversible if spontaneous pubertal development has not occurred or been induced. This was shown in the above patient by the return to subnormal growth hormone responses at a time when the plasma testosterone returned to an early pubertal level. This observation has not been reported pre-

viously. Third, when growth hormone reserve is questionable, the use of "testosterone priming" remains a useful diagnostic tool. However, in contrast to previous studies, it appears that a dosage as low as 100 milligrams of long-acting testosterone as a single injection may be adequate to enhance the growth hormone response. Fourth, in view of the precautions that must be taken when inducing hypoglycemia with insulin and the possible discomfort to the patient during the test (10), it may be adequate to test these boys with L-DOPA alone. In the event of an inadequate or equivocal response, repeating the L-DOPA stimulation after a single low-dose injection of testosterone may provide a more simple and convenient means to assess growth hormone reserve. However, additional cases should be studied before this priming method can be recommended as standard procedure. Finally, although the above data indicate again a correlation between testosterone and growth hormone secretion, the exact relationship of the pubertal growth spurt to the sex, tropic, growth and possibly other hormones or factors remains to be elucidated.

Acknowledgments

We would like to thank Dr. Frank Ross for the referral of patient E.C., and Mrs. Erica Sapira and Ms. Linda Aronoff of

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The Role of Radionuclide Imaging in Metastatic Disease: The Mount Sinai Experience

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Abstract

All patients referred for bone scanning during a three-month period were included in this study. The referral indications, clinical information, and bone scans were reviewed. Of the 463 patients, 387 (83%) were referred for possible neoplasms, and the results of bone, liver, brain, and gallium scanning were compared. The data suggested several general strategies currently employed in working-up patients suspected of malignant disease. The utility of routine liver scans was established, while brain scanning was found not to be helpful in the absence of neurologic signs. While bone pain was found significantly more often in patients with metastases, it was only present in 6% of such lesions. The serum alkaline phosphatase was not a sensitive or specific indicator of bone or liver metastases.

Introduction

All service organizations should regularly survey their product utilization to determine patterns that might influence future use. Radionuclide imaging of bone is a sensitive diagnostic procedure for the detection of skeletal neoplasm and infection (1). The procedure has been usefully applied for many years and is currently the most frequently requested nuclear medicine investigation.

This study reviews the utilization of bone scanning by referring physicians, the significance of bone pain as a symptom of skeletal metastases, the utility of multiple screening scans, and the role of serum alkaline phosphatase as an indicator of metastases.

Materials and Methods

During a three-month period in 1977, 486 patients were referred to the Department of Physics/Nuclear Medicine for bone scans. This population was used to study the utilization of nuclear medicine in neoplastic disease and 463 scan files were located

and reviewed. The patient's name, hospital number, age, sex, number of bone scans, total number of scans, and an assessment of the information supplied with the referral note were tabulated. The referral forms provide space for the provisional diagnosis, the results of previous radionuclide and radiologic studies, surgical findings, and current medication. Space is also provided for patient history, physical examination findings, and the information desired from the scan procedure. Information supplied with the referral form was classified as None, if no information was provided in the history and examination space; Relevant, if sufficient information was given to help interpret the images; and Insufficient, if the information was irrelevant to the scan purpose (for example, neurologic findings in bone scan request). The presence of bone pain was noted and the site tabulated. The final diagnosis was established by histologic, surgical, and other investigational results.

Scans were reviewed to determine the presence of focal or diffuse skeletal disease and the frequency distribution of abnormalities was listed. An etiology was ascribed when sufficient ancillary information was available. The categories included: metabolic, metastatic, infective, degenerative, traumatic, Paget's disease, indeterminate, or combinations of these. Any available previous bone scans were reviewed for comparison with the present study.

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Brain, liver, and gallium scans, when available, were reviewed for evidence of metastatic disease.

Data was analyzed by chi-square analysis of contingency tables by the Statistical Analysis System (SAS) at the university computer center of The City University of New York using an IBM 360. The null hypothesis indicating no difference in the compared sample populations was used with the level of significance selected at 0.05.

Results

The 463 patient studies reviewed represented 20% of the total departmental work load during the period analyzed. The provisional diagnosis supplied on the referral form indicated that the scan was considered important or necessary in the work-up of neoplastic disease in 387 instances (83%), osteomyelitis (5%), x-ray abnormality (5%), clinical symptoms (4%), and abnormal laboratory results (3%). Of the total population, 366 (79%) had a definitive diagnosis established, and 318 (69%) a final diagnosis of malignant neoplasm.

The mean patient age was 60 years and 65% were female. The population was divided into first visits (35%) and follow-up (65%). The overall mean number of bone scans per patient was 1.48 (range 1-7), and of total scans 3.33 (range 1-19). The mean number of total scans obtained on patients undergoing initial diagnostic work-up was 2.88 (range 1-7), indicating that the conventional three-scan survey (bone, liver, and brain) was the practice. Those follow-up patients with more than one bone scan (mean 2.79, range 2-7) in their file record were analyzed, and the mean interval between scans was 6.2 months.

The bone scan was normal in 59% of the study population, the remainder had either diffuse or focal abnormalities. Twenty studies were diffusely abnormal, seven because of metabolic diseases (osteomalacia, hyperparathyroidism, myeloid metaplasia, etc.), and thirteen from malignant causes (including hypertrophic pulmonary osteoarthropathy, diffuse metastases, or associated hypercalcemia). The focal abnormalities were attributed to metastases in 76 instances, benign etiologies in 66 instances, and in 30 cases, the etiology was either unknown or combinations existed.

The clinical information supplied with the referral request was also analyzed. In only one instance was a provisional diagnosis not provided. Overall, the information supplied was considered sufficient in 60%, insufficient (or irrelevant) in 19%, and none in 21%. These groups were then analyzed to deter-

mine whether the information supplied varied with diagnostic certainty. When the diagnosis was established (that is, final), 24% of referrals supplied no additional information and this fell to 9% when the diagnosis was merely provisional. Table I demonstrates this significant ($p = 0.00001$) increase in information supplied when the diagnosis was uncertain (provisional).

The specific information extracted from the referral request form included the presence of bone pain and neurologic symptoms. The site of bone pain was tabulated for the entire population and the scan checked for a focal abnormality in that region. In 148 patients, 167 sites (range 1-3 per patient) were listed and 81 (48%) had an associated scan abnormality. Of the patients with proven tumor, there were 82 symptomatic sites and 45 (55%) had an associated abnormality, a similar result to the nontumor population ($p = 0.11$). In the metastatic subgroup, 35 patients had 45 symptomatic sites of which 31 (69%) had an associated scan abnormality ($p = 0.001$). However, these 45 sites represented a mere 12% of the symptomatic patients' lesions and 6% of the 726 total metastatic abnormalities identified. The symptomatic patients had a similar burden of lesions to the total metastatic group (8.13 versus 9.5 lesions/patient).

The serum alkaline phosphatase was correlated with the number of skeletal metastatic lesions in the 27 patients with serum values obtained within one month of the scan. While there was a linear correlation ($r = 0.73$), ten patients (37%) were within the normal range for this laboratory. In those patients with abnormal liver scans, the serum alkaline phosphatase (SAP) was elevated in eight of nine patients with skeletal metastases, not significantly different from the nine of 17 patients with proven tumor without skeletal metastases ($p = 0.067$). In the patients with tumor and no skeletal metastases, an elevated SAP provided no predictive value as to whether the liver scan was normal or abnormal.

The most current bone and other scans were correlated to determine whether other scanning procedures yielded evidence of metastases else-

TABLE I
Contingency Table of Diagnostic Certainty Versus
Information Quality

Stage of Diagnosis	Quality of Information Supplied (# patients)		
	None	Irrelevant	Sufficient
Final	84	51	212
Provisional	10	36	60

Chi-square = 24.5, df = 2, $p = .00001$.

where. Brain scans were performed on 162 patients (50% of patients with proven neoplasm) and Table II compares the scan result with the presence of focal or diffuse neurological signs as listed on the referral request. Scan abnormalities were not seen in the absence of stated focal neurological signs.

Gallium scans were obtained in 70 patients (22% of patients with proven tumor) and 32 scans demonstrated lesions, of which 11 were skeletal. Most of the abnormalities were seen in two tumor types. In three malignant melanoma patients, the skeletal abnormalities were identified equally well with bone and gallium scans. The 19 lymphoma patients with bone and gallium scans demonstrated lesions equally well in 17 and, in two instances, skeletal abnormalities were not identified with gallium.

Three hundred and twelve patients had a liver scan performed in addition to a bone scan, and this represented 67% of the patients with proven tumor. The liver scans were classified as normal variants (71%), or abnormal with either focal (10%) or diffuse (19%) patterns. In the proven tumor patients the overall incidence of focal hepatic abnormalities, and thus metastatic disease, was 11%. The subset of the largest tumor groups (breast, lung, and gastrointestinal) with both scanning procedures was further evaluated (Table III). While the incidence of skeletal metastases ($p = 0.81$) and diffuse hepatic disease ($p = 0.68$) was similar in each group, the incidence of focal abnormalities on liver scanning was significantly different ($p = 0.01$). Analysis of these tumors determined that this difference was due solely to the higher incidence in gastrointestinal (GIT) tumors ($p = 0.001$).

The data were analyzed to determine whether the presence/absence of skeletal metastases affected the incidence of liver scan abnormalities. The presence of skeletal metastases did not alter the overall incidence of focal ($p = 0.84$) or diffuse (0.70) liver scan abnormalities. In breast ($p = 0.99$), lung ($p = 0.68$), and GIT ($p = 0.16$) tumors, no significant difference in the rate of detected liver scan abnormalities was found based on the presence/absence of skeletal metastases.

Discussion

The purpose of an analytical review such as this is to review current practices, reinforce efficacious ones, and propose modifications.

The data indicates that the majority of patients undergoing bone imaging (83%) are referred for workup or follow-up of tumor and that they fre-

TABLE II
Presence of Neurologic Signs on Referral Form Versus Presence of Brain Scan Abnormality

Neurological Signs	Scan Result	
	Normal	Focal Lesion Present
None	136	0
Focal	7	13
Diffuse	6	0

TABLE III
The Incidence of Bone Scans Demonstrating Metastatic Disease and Liver Scan Abnormalities in the Major Tumor Categories

Region	# of Patients	# of Abnormal Bone Scans (%) ^a	# of Abnormal Liver Scans (%)	
			Focal	Diffuse ^c
Breast	71	19 (27%)	4 (6%)	18 (15%)
Lung	50	16 (32%)	7 (14%)	5 (18%)
Gastrointestinal tract (GIT)	29	8 (27%)	11 (34%)	4 (14%)

^a Chi-square = 0.41, df = 2, $p = 0.81$.

^b Chi-square = 10.2, df = 2, $p = 0.01$.

^c Chi-square = 0.77, df = 2, $p = 0.68$.

quently have a three scan workup (bone, brain, and liver scan). Repeat or follow-up bone scans in these patients follow a pattern of six monthly intervals.

In 41% of the total patient population, the bone scans were abnormal and 40% of these abnormalities were attributed to metastases. The clinical information supplied with the bone scan request was significantly more complete when the patient's diagnosis was uncertain. This increase in clinical detail is helpful to the nuclear medicine physician in reading the scans and therefore probably results in better reports to the referring physician.

Much has been made of the usefulness of bone pain in metastatic disease. In this series, 69% of the patients with metastases and bone pain had scan abnormalities at the symptomatic site(s), which corresponds well with reports of 56%, 58%, and 65% in other series (2-4). However, only 6% of all metastatic lesions were painful at the time of scanning.

The serum alkaline phosphatase results confirm other reports of being related to the number of skeletal metastases (5). When this is elevated in a tumor patient without skeletal metastases, there is an equal chance of the patient having a normal, focally abnormal, or diffusely abnormal liver scan. These data confirm the serum alkaline phosphatase as an insensitive nonspecific indicator of hepatic and skeletal metastases.

With respect to other nuclear medicine procedures as part of the workup of tumor patients, the data indicate that brain scans are unwarranted as

a screening procedure in the absence of neurologic signs. This confirms other reports (6, 7), although up to 5% of patients with carcinoma of the lung may have cerebral metastases without neurologic signs (8-10). Gallium scanning is particularly useful in staging lymphoma and carcinoma of the lung (11, 12), and in our experience, it is also useful in malignant melanoma when soft tissue and skeletal spread is identified by this single imaging procedure (13).

The utility of the liver scan in addition to the bone scan is confirmed by this study. While the overall incidence of hepatic metastases in the entire tumor group is relatively low (11%), this incidence is variable and significantly different in GIT tumors (34%) when compared to the other tumor groups. This difference might be expected as a result of the portal drainage of much of the GIT and the interposition of liver between the primary tumor and the systemic circulation. In the major tumor categories (breast, lung, and GIT) the incidence of hepatic metastases was examined according to whether skeletal metastases were present or not. It might have been expected that metastases present in one organ would result in an increased incidence in another system, especially when hematogenous spread was common to both organ systems. However, the data refutes a relationship between the presence of skeletal metastases and the occurrence of hepatic metastases. This reinforces the use of routine hepatic imaging regardless of the result of the bone scan in tumor patients.

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Sarcoid-like Granulomas in Secondary Oxalosis: A Case Report

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Abstract

The autopsy of a 47-year-old woman with chronic renal failure maintained on hemodialysis for five years showed severe secondary oxalosis, manifested in the lymph nodes, spleen and liver as sarcoid-like epithelioid granulomas. Though seemingly an unusual morphologic response to oxalate deposits, this lesion appears to be a new addition to the entities to be considered in the differential diagnosis of non-necrotizing epithelioid granulomas. The nature of this lesion is discussed.

Secondary oxalosis in chronic renal failure is well documented (1, 2). Organs that are involved include the kidneys (1, 2), myocardium (3), thyroid (1, 4), less commonly the spleen (1), lungs (1), and rarely lymph nodes. Even though the functional sequelae of oxalate deposit have been stressed in some papers (3, 6), less attention has been paid to the pathologic response of the tissues and the structural changes induced by oxalate in various organs. Most commonly mentioned alterations are fibrosis (1, 3, 6) and foreign body granulomas (1, 4, 6). In an autopsy study of patients maintained on hemodialysis, extensive oxalate deposit associated with sarcoid-like granulomas were found in the spleen, lymph nodes and liver of a patient. The present communication describes this case and discusses the significance of this lesion.

Case Report

A 47-year-old white woman with known end-stage kidney disease maintained on hemodialysis for five years was admitted because of fever, intermittent confusion, muscular twitching and nuchal rigidity. She was discharged from the hospital a few weeks earlier with a diagnosis of uremic encephalopathy. At admission blood pressure was 180/100 and her temperature 102°F. Laboratory data were as follows: Sodium 135 mEq/dl, potassium 5.0 mEq/

dl, chloride 92 mEq/dl, bicarbonate 9 mEq/dl, blood glucose 101 mg/dl. The hospital course was dominated by neurologic symptoms and signs consisting of repeated seizures, confusion, occasional stupor and lethargy and paranoid tendencies. The electroencephalogram showed moderate to severe diffuse cortical dysfunction. Terminally, she developed left hemiparesis progressing to hemiplegia. She was dialyzed numerous times and was given supportive therapy. She expired on the eighth hospital day.

Postmortem Examination

Gross

The heart weighed 450 grams. There was left ventricular hypertrophy. The valves were unremarkable. Coronary atherosclerosis was moderate.

The lymph nodes, particularly in the abdominal group, were enlarged the largest measured 3.5 cm in maximum dimension. They were moderately firm and showed pin head size whitish-gray nodules on cut section.

The spleen weighed 400 grams and displayed a smooth slaty gray capsule. It was firm and cut section revealed whitish-gray nodules ranging in size to 2 cm in maximum dimension. Both kidneys weighed 55 grams. The cortical surface was finely granular and section showed loss of corticomedullary distinction. The intrarenal branches of the renal artery were patent and prominent. A few

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cortical cysts were observed. The pelvicalyceal system was unremarkable.

The brain showed diffuse cerebral edema. Focal lesions were absent.

Microscopic

The kidneys showed extensive deposition of oxalate crystals in tubules and interstitium. The crystals were identified as previously described (1). Occasional foreign body granulomata were seen. The primary renal disease was chronic glomerulonephritis.

The architecture of the lymph nodes was generally obliterated by multiple non-necrotizing epithelioid granulomas evenly scattered in the cortex and the medulla (Fig 1). Calcium oxalate crystals, identified

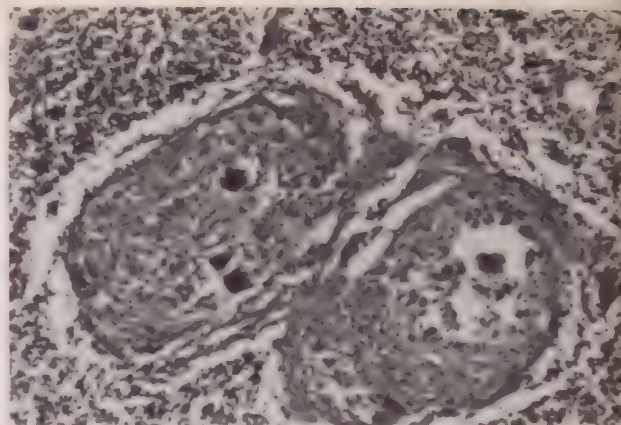


FIG. 3. High power detail of two granulomas with epithelioid cells and considerable fibrosis (H & E, $\times 100$).

as previously described, were commonly found in nearly all granulomas, some located within giant cells (Fig 2). Both Langhans and foreign body type giant cells were present (Fig 3). Focal calcification was occasionally observed.

Granulomas of larger sizes, similar to those in the lymph nodes were present in the spleen. In other areas, however, hyalinized fibrosis containing oxalate crystals were seen. There was considerable hemosiderosis.

The lobular architecture of the liver was undisturbed. There was a random distribution of granulomas and hyalinized areas containing oxalate crystals. Other salient findings were cerebral edema, bronchopneumonia, and colonic diverticulosis.

Discussion

Considering that calcium oxalate is inert, the mechanism by which it evokes tissue reaction is not understood. It has been suggested that dissociation of this compound with liberation of toxic oxalate ions may be the basis of the tissue injury (7). The types of tissue reaction that have been described include necrosis (8), chronic inflammation (8), fibrosis (1-3), and granulomatous reaction (1, 9). In many cases, the presence of oxalate in tissue excites no reaction whatever (1).

Deposition of oxalate in lymph nodes is extremely rare. Involvement of lymph nodes was not encountered in two large autopsy series of patients with chronic renal failure (1, 2). In the case reported by Klauwers et al, in a 22-year-old man with failure of renal transplantation, oxalate deposition was found in lymph nodes in addition to numerous other organs (5). Further details of the histologic features of the lymph nodes were not given. Dystrophic oxalosis was described in the lymph node of a 54-



FIG. 1. Obliteration of lymph node architecture by granulomas (G) of varying sizes, some showing fibrosis. Scant remnants of native lymphoid tissue (L) are present. (H & E, $\times 18$).

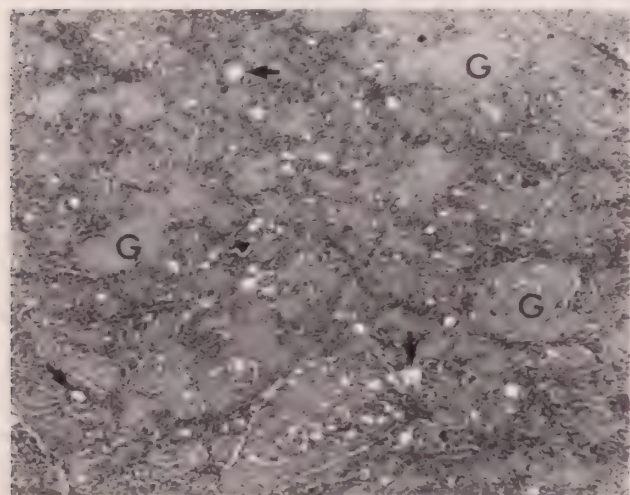


FIG. 2. Lymph node showing extensive oxalate deposit (arrows) within and outside sarcoid-like granulomas (G). (H & E, semipolarized light, $\times 40$).

year-old woman with a presumptive diagnosis of sarcoidosis (10). Noncaseating granulomas were present also in the spleen and liver.

The latter case raises the question as to whether our patient had chronic renal failure with secondary oxalosis or a combination of chronic renal failure and sarcoidosis with dystrophic oxalosis. The presence of noncaseating granulomas in the liver, lymph nodes and spleen would suggest a diagnosis of sarcoidosis. The clinical and laboratory features of sarcoidosis, however, were not observed in the patient. Furthermore, the absence of hilar lymphadenopathy, and pulmonary involvement do not favor a diagnosis of sarcoidosis. Unfortunately Kveim test was not performed. That the granulomatous reaction is secondary to oxalate deposit is supported by the following: 1) calcium oxalate crystals were present, sometimes in abundant quantity within virtually all the granulomas in the various organs, 2) there was severe renal oxalosis in the absence of a granulomatous reaction, and 3) the spleen and the liver showed nonspecific fibrosis within which oxalate crystal were demonstrable. The granulomatous reaction evoked by oxalate deposits previously described by us (1) and others (9) has been of the foreign body type; to our knowledge, sarcoid-like granulomatous response has not been reported.

The pathogenesis of these granulomas is unclear. We suggest that they probably result from an idiosyncratic (hypersensitivity) reaction to calcium oxalate. This conclusion is also supported by the rarity of this reaction and the morphologic similarity to granulomas known to have a hypersensitivity basis

such as brucellosis, berylliosis and certain drug-induced liver granulomas. It is important to recognize this entity and distinguish it from sarcoidosis. The diagnostic and therapeutic implication of this distinction is evident and cannot be over emphasized.

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Seborrheic Keratosis with Epidermal Cyst

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The majority of keratin-containing skin cysts arise from various portions of the pilosebaceous apparatus. Those that arise from the infundibulum are designated epidermal cysts. Although it is well known that epidermal cysts occur not uncommonly in association with nevocellular nevi, it seems to have escaped medical attention that epidermal cysts also accompany seborrheic keratoses. I have observed twelve cases during a period of eight consecutive months representing approximately 0.5% of all seborrheic keratoses examined during that period. This represents the subject of my report.

Materials and Methods

Twelve lesions were biopsied from twelve patients and were fixed in Bouin's solution, routinely

processed, embedded in paraffin and stained with hematoxylin and eosin.

The clinical data are summarized in Table I. All lesions had been present for many years and had shown recent enlargement. Histologically, all cases contained a large keratin-containing cyst lined by focally attenuated stratified squamous epithelium beneath a seborrheic keratosis (Figs. 1, 2) of either the acanthotic (nine cases) or reticulated (three

TABLE I

Seborrheic Keratosis with Epidermal Cyst

Case	Age	Sex	Location	Size*	Clinical Impression
1	74	F	Chest	0.7 cm	Epidermal cyst
2	58	M	Upper eyelid	0.5 cm	Papilloma
3	56	M	Groin	0.6 cm	Cellular nevus
4	76	M	Lower eyelid	0.5 cm	Epidermal cyst
5	64	F	Temple	0.4 cm	Seborrheic keratosis
6	68	M	Lower eyelid	0.7 cm	Epidermal cyst
7	89	M	Cheek	0.8 cm	Epidermal cyst
8	54	M	Nose	0.4 cm	Cellular nevus
9	70	M	Cheek	0.5 cm	Epidermal cyst
10	50	M	Neck	0.4 cm	Cellular nevus
11	57	F	Neck	1.0 cm	Epidermal cyst
12	62	M	Forehead	0.7 cm	Seborrheic keratosis

* Size represents maximum dimension.

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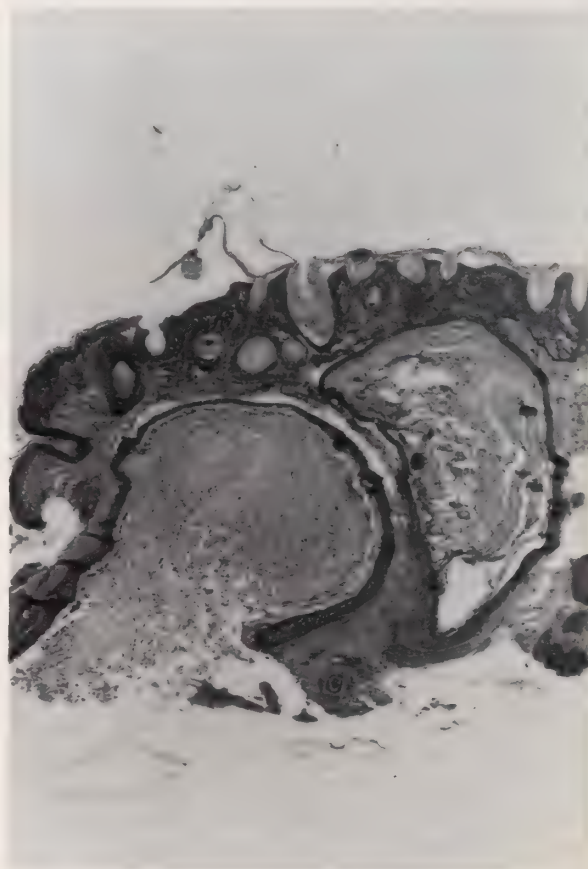


FIG. 1. Large cyst deep to and connected to overlying seborrheic keratosis of the reticulated type in Case I (H & E $\times 20$).

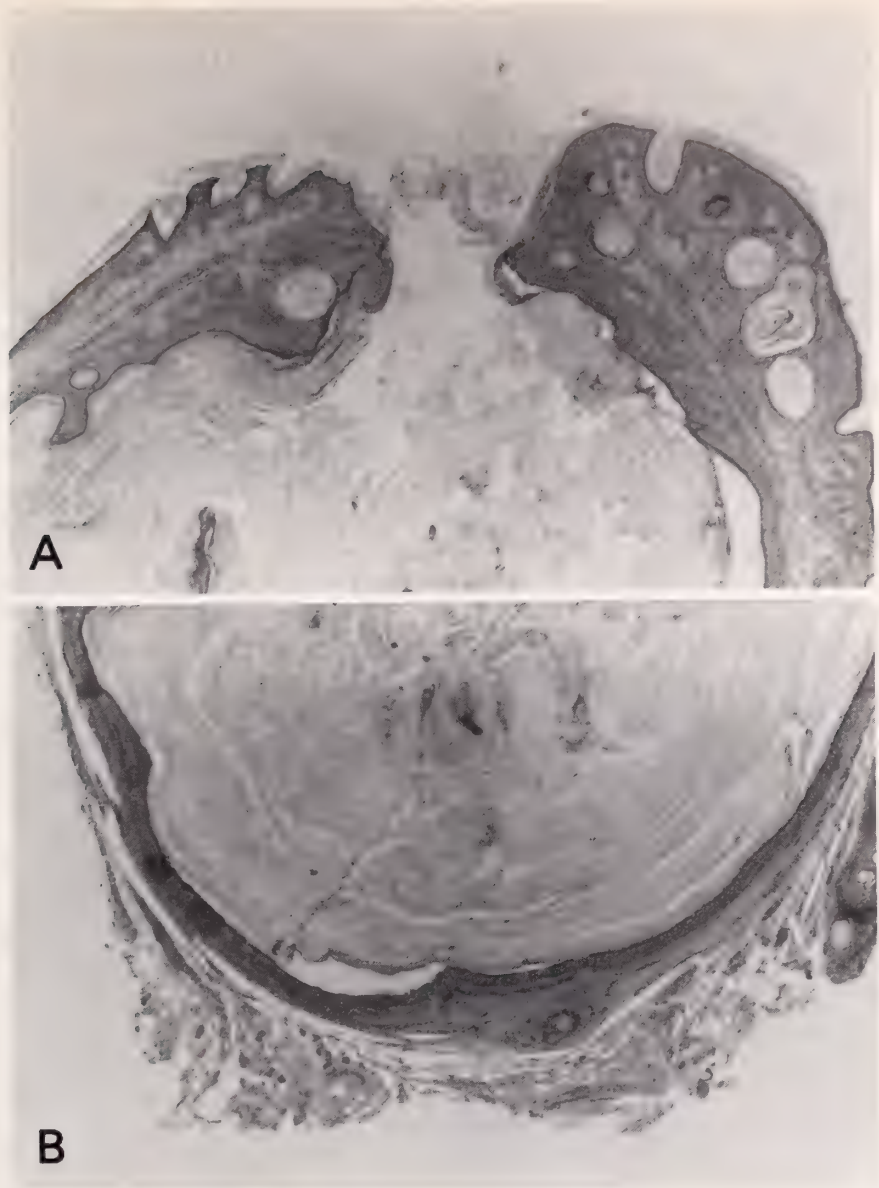


FIG. 2. Large cyst deep to and communicating with skin surface of overlying seborrheic keratosis of the acanthotic type in Case II. A and B represent top and bottom half of the cyst, respectively (H & E $\times 20$).

cases) type. Portions of hair shafts were also observed within the lumens in two cases. Except for three cases, all of the remaining cysts (nine cases) were connected to the overlying seborrheic keratosis, five of them communicating with the skin surface. The stroma about all lesions was focally edematous and infiltrated with inflammatory cells consisting of lymphocytes, histiocytes and neutrophils as well as melanophages in nine cases.

Discussion

The histopathology of the various types of seborrheic keratoses is well documented (1) and need not

be detailed here. The pathogenesis of these lesions is still controversial. Suffice it to say that seborrheic keratoses represent delayed epithelial nevi composed of keratinocytes having a somewhat altered metabolism. In addition to the presence of both pseudocysts and bona fide cysts filled with keratin, dilated sweat ducts and hair follicles plugged with keratin often traverse seborrheic keratoses. It should therefore be expected for epidermal cysts to develop because of obstruction to the intralesional portion of the pilosebaceous duct by the proliferating adjacent keratinocytes of the seborrheic keratosis. What is surprising is that this fairly common association does not seem to have received much,

if any, attention in the medical literature. The pathogenesis of epidermal cysts with nevocellular nevi is analogous and this association is well known. It is interesting that, in my series, these cysts occurred with the acanthotic and reticulated types and were not observed in the hyperkeratotic type of seborrheic keratosis. They tended to be located about the head and neck in the elderly and showed a male predominance. This combination of lesions represents yet another histologic alteration that can be added to the list of those occurring in the ever changing seborrheic keratosis (2).

Summary

Twelve seborrheic keratoses associated with large epidermal cysts are reported, representing approx-

imately 0.5 percent of all seborrheic keratoses examined. The pathogenesis of these cysts is most likely pilosebaceous duct obstruction by the proliferating keratinocytes of the seborrheic keratosis about the traversing duct. This combination of lesions can be added to the list of histologic alterations that can take place in the ever changing seborrheic keratosis.

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Functional Interrelationships of Exocrine Pancreas and Endocrine Glands in Health and Disease

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Information about the effects of the "classical" hormones, and of diseases of the hormone-producing endocrine glands, on the functions of the exocrine pancreas is scanty, patchy and discordant—which is surprising in view of the powerful regulatory effects of these hormones on visceral function in general. Worse, we know that blood-borne hormonal peptides can exert very specific pancreatic regulatory effects, but our knowledge depends on inferences from the pancreatic effects of invariably pharmacologic doses of exogenous peptides or on the results of pathologically excessive or defective secretion of individual hormones in disease. Consequently, we know little or nothing about the physiologic effects and interactions of these hormones with each other, with the gut peptides, and with the neural control of pancreatic exocrine function.

This review is intended to outline the paucity of our information in the hope of stimulating further studies of this potentially important topic.

Parathyroid Hormone

Effect of Parathyroid Hormone on the Exocrine Pancreas

No systematic studies have been carried out on the effects of parathyroid hormone on the human pancreas. In rats with transplanted parathyroid glands, the greater the amount of transplanted parathyroid tissue, the greater the incidence and

degree of pancreatitis (1). It was concluded from this study that the parathyroid glands increased serum calcium levels which, in turn, increased the amounts of calcium secreted in pancreatic juice, resulting in activation of trypsinogen to trypsin and producing pancreatitis. This sequence of events has not been confirmed in the causation of human pancreatitis.

Hyperparathyroidism

In 1957, Cope (2) reported that the association between hyperparathyroidism and pancreatic disease represented a significant causal relationship. Since then, many reports have confirmed the considerable coincidence of pancreatic disease and hyperparathyroidism (3, 4, 6-9). The pancreatic disease can manifest as acute pancreatitis (3, 8, 10-12) or may manifest as chronic pancreatitis with clinical evidence of severe exocrine insufficiency and often pancreatic calcification (6, 13, 14). In patients with chronic hyperparathyroidism, enzyme output was impaired while bicarbonate secretion remained normal (15).

Most of the mechanisms that have been proposed to explain how hyperparathyroidism produces pancreatitis involve a hypothetical noxious effect of increased amounts of ionized calcium in the extracellular environment of the pancreatic cells. For example, it has been proposed that excess of parathyroid hormone produces pancreatic necrosis (10, 16) either directly or because increased amounts of calcium ions accelerate the autocatalytic activation of trypsinogen by trypsin and inhibit the self-inactivation of trypsin (17). It has also been suggested that there is precipitation of calcium salts within the alkaline luminal content of the pancreatic duct (2, 7, 10, 18), with obstruction to flow and conse-

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quent pancreatic damage, or perhaps release of pancreatic proteases into the circulation, with resultant formation of pancreatic arterial thrombi and pancreatic infarction (11, 12, 19).

Although occasional reports mention that the patients with hyperparathyroidism who develop acute pancreatitis have entirely normal serum calcium levels (20), it seems possible that the principal effect of hyperparathyroidism on the exocrine pancreas is not a direct one, since the hypercalcaemia caused by hypervitaminosis D (21), multiple myeloma (22), breast carcinoma with metastases (23), and the hypercalcaemia following calcium infusion (24) has also been reported to cause acute pancreatitis. The apparent importance of the concentration of circulating ionized calcium in the pathogenesis of pancreatic disease, the influence of hyper- and hypocalcaemia on pancreatic function has been studied both in animals and man. Studies on healthy subjects showed that acute intravenous administration of calcium salts significantly increased the concentration and output of pancreatic enzymes, without affecting the secretion of bicarbonate and water (25, 26). These results were observed both under basal conditions and during stimulation with secretin and cholecystokinin (CCK), but not with CCK alone (25). The mechanism of the stimulant effect of calcium on pancreatic enzyme secretion has not been defined and a number of suggestions have therefore been made. For example, it has been suggested that calcium induces release of transmitter substances from the vagal nerve endings, as shown by study of the perfused pancreas (27). It seems probable that calcium also exerts a direct action on the pancreatic acinar cells, by stimulating exocytosis (28), and by influencing the formation and activation of cyclic AMP and GMP, that are involved in the process of enzyme secretion (29). In addition, calcium stimulates the release of endogenous gastrointestinal hormones like gastrin (30) and may, therefore, also influence the release of the hormones from the gut.

Whatever the effects of calcium on pancreatic enzyme secretion, these findings do not, of course, shed any light on the apparently damaging effects of calcium on the intact pancreas.

Hypoparathyroidism

No satisfactory reports of the effect of hypoparathyroidism on the exocrine pancreas are available. Experimental hypocalcaemia inhibits the pancreatic secretory response to stimulants both in man (26) and in vitro (27).

Effects of Pancreatic Disease on Parathyroid Function

The converse relationship, of the pancreas on parathyroid function and serum calcium levels, is also well recognized. Thus, acute pancreatitis is often associated with severe hypocalcaemia (31-33) the cause of which has given rise to much controversy. On the one hand, it has been reported that parathyroid hormone may be undetectable in patients suffering from acute pancreatitis (34) or significantly less than normal (33, 35). The administration of calcium salts has been reported not to improve the hypocalcaemia, while the administration of parathyroid hormone is followed by return of serum calcium levels to normal, resulting in disappearance of symptoms such as tetany (32, 36). The absence of parathyroid hormone has been considered to reflect parathyroid failure or exhaustion (34), while low levels have been thought to indicate an inadequate response (37). The therapeutic use of parathyroid hormone for the treatment of hypocalcaemia and hypomagnesaemia occurring early in the course of acute pancreatitis has been recommended (38).

Since it has been reported that the concentrations of parathyroid hormone are normal or greater than normal in acute pancreatitis with hypocalcaemia (34, 35, 39), resistance to the action of the hormone (37) has also been proposed to account for the hypocalcaemia.

It seems clear, therefore, that whatever the effect on the parathyroid glands, pancreatic disease exerts quite profound effects on serum calcium levels in patients with acute pancreatitis. Similarly, the steatorrhoea resulting from chronic pancreatitis exerts a hypocalcaemic effect which is, of course, more continuous than the transient effect of acute pancreatitis. Thus, the parathyroid disease that is associated with chronic pancreatitis may be secondary to the pancreatic disease, rather than primary and the cause of the pancreatic disease, at least in some patients (13, 40, 41). That is, the hyperparathyroidism is initially a functional response to repeated, or continuous, hypocalcaemia (41) and may, in due course, become autonomous as in other types of "tertiary" hyperparathyroidism (42).

Calcitonin

Effect of Calcitonin on the Exocrine Pancreas

In man (26, 43), cats (44), dogs (45) and isolated canine pancreas (46) calcitonin inhibits hormonally stimulated pancreatic secretion. While in the cat

and dog this inhibition involves the secretion both of enzymes and bicarbonate, in man calcitonin infusion results in a significant reduction of the enzyme response to secretin and CCK, without change in bicarbonate output or secretory rate (26, 47, 48). No changes in serum calcium levels were detected during the inhibition (26) and calcium infusion did not alter the inhibitory response to calcitonin (47), suggesting that calcitonin exerts a direct effect on the pancreatic acinar cells, without involving ionic calcium. Purportedly, calcitonin inhibits pancreatic secretion by increasing the movement of calcium ions from the intracellular to the extracellular space. This results in depletion of the intracellular calcium that is necessary for the secretion of enzymes (49). In this connection, it has been shown by perfusing animal pancreases *in vitro* with fluid containing different concentrations of calcium that the pancreatic secretion of enzymes depends on the intracellular, as well as extracellular, ionic calcium (27, 50).

To add the usual note of discord, it was reported that in both man and dog, calcitonin failed to inhibit pancreatic exocrine secretion and, indeed, produced slight stimulation of human bicarbonate secretion (51).

Hypercalcitoninaemia

A study of pancreatic exocrine secretion has been carried out in two patients with medullary carcinoma of the thyroid with hypercalcitoninaemia, as well as in patients with Paget's disease of bone during treatment with calcitonin. Pancreatic function was normal in the tested individuals, suggesting that the pancreatic exocrine cells had adapted to the inhibitory effects of chronically elevated levels of circulating calcitonin (52).

Effects of Pancreatic Disease on Calcitonin Levels

Circulating levels of calcitonin have been found to be abnormal in pancreatic disease. For example, hypercalcitoninaemia has been described in patients with acute pancreatitis and assumed to play some part in the pathogenesis of the hypocalcaemia characterising the acute attacks (37, 53–55). The mechanism of the hypercalcitoninaemia has not been defined, but it has been suggested that the hyperglucagonaemia that also occurs in acute pancreatitis may play some part, since glucagon stimulates calcitonin release (56). It has also been suggested that gastrin is involved in the hypercalcitoninaemia of acute pancreatitis (53) since gastrin is a

powerful stimulant of calcitonin release (57). However, blood levels of gastrin have been reported to be normal in acute pancreatitis (58).

There have, of course, also been reports of normal, or undetectable, calcitonin levels in patients with acute pancreatitis (34, 53, 58).

Thyroid Hormones

Effects of Thyroxine on the Exocrine Pancreas

The effects of thyroxine on human exocrine pancreatic secretion have not been systematically studied. In rats with thyroxine-induced hyperthyroidism, increase in the rate of secretion of pancreatic juice and in output of enzymes has been demonstrated, compared with untreated animals (59). Prolonged administration of thyroxine produced an increase in the weight of the pancreas and in enzyme content (59). Thyroxine administration to hypophysectomised rats increased the synthesis and secretion of pancreatic enzymes (60).

Hyperthyroidism

Human pancreatic function has been little studied in hyperthyroidism. The pancreatic exocrine secretory response was considered normal in some patients with steatorrhoea attributed to thyrotoxicosis but the testing of pancreatic function was unsatisfactory and enzyme secretion was not measured (61). It has, however, been reported that the pancreatic response to a meal is about half that in the euthyroid state during active hyperthyroidism (62).

A case of acute pancreatitis in a patient with thyrotoxicosis and hypercalcaemia has been reported (63).

Hypothyroidism

No systematic investigation of pancreatic exocrine function has been performed in patients with hypothyroidism. In dogs, thyroidectomy reduced all the indices of basal secretion, including secretory rate, bicarbonate secretion and enzyme production but only the enzyme response to hormonal stimulation with CCK and secretin was reduced (64). Similar results have been reported in rats following thyroidectomy (65). The mechanism by which thyroid deficiency affects pancreatic secretion has not been defined, but it has been speculated that the reduced secretion of pancreatic enzymes reflects absence of the anabolic effect of thyroxine, since the latter hormone stimulates the incorporation of amino acids into proteins (66).

Pancreatic exocrine insufficiency has been reported in a patient with hypothyroidism (and Klinefelter's syndrome) (67). In patients suffering from hypothermia associated with hypothyroidism, raised concentrations of serum amylase have been reported and attributed to acute pancreatitis (68).

Effect of Pancreatic Disease on Thyroxine Metabolism

In patients with pancreatic steatorrhoea, the fractional rates of turnover of thyroxine, as well as total daily degradation and faecal losses were greater than normal. The net peripheral tissue turnover was normal (69).

Thyrotropin-releasing Hormone

Although TRH appears to be present in the pancreas (70), the hormone has no effect on basal or stimulated pancreatic secretion (71).

Anterior Pituitary Hormones

Effects of Anterior Pituitary Hormones on the Exocrine Pancreas

Surprisingly little has been written about the effects of the anterior pituitary hormones on exocrine pancreatic function. Indeed, only prolactin and ACTH appear to have been studied.

In dogs, intravenous administration of prolactin inhibited pancreatic secretory rate and bicarbonate output, without affecting enzyme secretion (72). In man, ACTH reduced the enzyme-secretory response to secretin and CCK, without affecting bicarbonate secretion (73, 74) although a different report indicates that bicarbonate secretion is also sometimes reduced (75).

Anterior Pituitary Hyperfunction

There have been no satisfactory studies of pancreatic exocrine function in the syndromes of anterior pituitary hyperfunction.

Anterior Pituitary Hypofunction

Experimental studies in animals showed that hypophysectomy resulted in morphologic (76, 77) and functional (78) features suggestive of impaired function of the exocrine pancreas. The histologic investigations demonstrated that reduction in pancreatic glandular weight (79) was associated with atrophy of the acinar cells and decrease in the intracellular zymogen granules, resulting in a significant decrease in pancreatic enzyme content (77, 79, 80). These

findings correlated well with the results of study of the secretin-stimulated pancreas (78) which showed a significant decrease in secretory rate and bicarbonate and enzyme outputs. The functional impairment of the exocrine pancreas after hypophysectomy appears to be caused by impaired adrenocortical function, since the administration of ACTH or corticosteroids produces complete (78) or partial (81) return to normal secretory function. Pancreatic function has not been systematically studied in human hypopituitarism although mention is made of pancreatic hypofunction in three patients with hypopituitarism (82).

Posterior Pituitary Hormones (Antidiuretic Hormone) (ADH)

Effects of ADH on the Exocrine Pancreas

Intravenous injections of increasing doses of an ADH preparation decreased the rate of secretion of pancreatic juice in response to secretin without change in the concentration of bicarbonate, while the concentrations of protein and amylase increased, so that the output of protein tended to remain constant, or increase, during ADH administration (83). Later studies showed that ADH produced a significant, but transient, reduction in secretory rate and bicarbonate concentration and output from the secretin-stimulated canine pancreas (84). The inhibitory effect of ADH on the canine pancreas has been confirmed and it has been demonstrated that there is a direct relationship between the degree of inhibition of the pancreatic response to secretin and the dose of ADH (85). The effect of ADH on human pancreatic secretion stimulated by continuous intravenous infusion of secretin was similar with significant decrease in secretory rate, bicarbonate concentration and output and a significant increase in the concentration of calcium, magnesium and total protein (86). There was rebound hypersecretion of pancreatic juice when the effects of the ADH injections wore off.

The mechanism by which ADH inhibits the exocrine pancreas has not been defined. A direct inhibitory effect on pancreatic secretion has been proposed (84). However, recently, an indirect effect—vasoconstriction—seems to have been established by use of two synthetic analogues of vasopressin, one with antidiuretic effects (which had no effect on the pancreatic secretion of bicarbonate) and one with pressor effects (which decreased pancreatic blood flow and the secretion of bicarbonate) (87).

No reports are available about the state of pan-

creatic exocrine function in diabetes insipidus nor in syndromes associated with abnormally great secretion of ADH.

Corticosteroids

Effects of Cortisone on the Exocrine Pancreas

Cortisone and derivatives were reported not to affect hormonally stimulated pancreatic function in man (72) while a study of patients treated with long-term steroids was interpreted as showing hypersecretion of pancreatic enzymes in the treated patients, compared with normal individuals (88). Similar hypersecretion of protein was reported in rats receiving long-term treatment with steroids (89). However, other studies showed inhibition of pancreatic secretion after intravenous injection of steroids during acute experiments in man (74) as well as after acute or prolonged infusion in dogs (90).

The apparent functional pancreatic changes produced by steroids have been considered to bear some relationship to the morphologic changes observed in the pancreas of animals treated with steroids. For example, steroid-treated rats developed dilated acini, flattened ductal epithelium and ductal protein plugs (89). In steroid-treated rabbits, the pancreatic acinar cells showed reduced basophilia, vacuolisation and nuclear abnormalities (91). Autopsies on patients treated with ACTH showed pancreatic ectasia, as well as acute pancreatitis of varying degrees of severity and focal necrosis of the peripancreatic fat (92).

The pancreatic morphologic changes following experimental administration of steroids have been assumed to relate, in some manner, to the acute pancreatitis that has been reported to occur occasionally in patients treated with steroids for some other disease. Steroid-associated pancreatitis has been described in children and also in adults and may accompany either increase or decrease in steroid dosage (93, 94).

Steroids have been described, in a number of reports, as exerting a quite different effect on the pancreas, that of being a growth or developmental factor when administered to nursing rats. In rats, hydrocortisone induces maturation of the acinar cells after birth, particularly when combined with CCK (95). Similarly, hydrocortisone increases the amylase activity of the pancreas in mice during postnatal development (96) and dexamethasone (and ACTH) increase the amylase activity of infant rats (97) while hydrocortisone stimulates the accumulation of chymotrypsinogen in pancreatic ex-

plants from chick embryos (98). A further analysis of the enzyme induction produced by steroids has shown that hydrocortisone prevents the decrease in pancreatic amylase and chymotrypsinogen that characterises the postnatal period in rats, the usual decrease perhaps attributable to the decreased pituitary and adrenal activity during the first weeks after birth (99).

No information is available about pancreatic exocrine function in Cushing's syndrome.

Adrenal Cortical Hypofunction

No information is available about exocrine pancreatic function in Addison's disease.

The effects of adrenalectomy on the rat pancreas have been studied with contradictory results (100–103). Following adrenalectomy, the pancreatic content of amylase was reported to be unchanged (100) while other studies (101) showed that the pancreatic content of amylase decreased significantly after adrenalectomy (102) and that secretion of enzymes and bicarbonate was impaired (103).

Adrenaline

Effects of Adrenaline on the Exocrine Pancreas

A number of studies have been reported of the effects of adrenergic hormones and drugs, and also of antagonists to these drugs, on exocrine pancreatic function. There are very marked species differences but, in general, adrenaline inhibits pancreatic secretion of enzymes and bicarbonate in intact animals (72, 104) and suppresses the discharge of zymogen granules from the pancreatic acinar cells (105). The phase of pancreatic inhibition produced by adrenaline may be succeeded by "rebound" hypersecretion (106). Adrenaline appears to be a more powerful inhibitor of pancreatic secretion than noradrenaline (106).

The mechanisms by which these hormones influence pancreatic secretion have not been defined. It has been proposed that adrenaline and noradrenaline directly inhibit pancreatic acinar cells (106, 107) although it has also been concluded that the pancreatic inhibition reflects the vasoconstriction caused by the hormones (106, 108). The vasoconstrictive effect, mediated by alpha-adrenoceptors, can be blocked by alpha-receptor blocking drugs without affecting the inhibitory actions of adrenaline (106). Moreover, the alpha-adrenergic inhibition of pancreatic secretion by noradrenaline can be converted to dose-dependent stimulation after administration of the alpha-adrenergic blocker

phenoxybenzamine, indicating that noradrenaline also has a direct stimulating effect on the pancreas (108, 109). The stimulant effect of noradrenaline on the pancreas has been considered to represent a weak dopaminergic action of the hormone (110). A pancreatic stimulant effect of adrenaline and noradrenaline has also been reported in anaesthetised intact rats (111). The effect of beta-adrenergic stimulation on pancreatic secretion is also controversial (104) but it seems that in the dog, beta-adrenergic stimulation inhibits pancreatic secretion (112, 113) without causing vasoconstriction (114). In the rat, beta-adrenergic drugs stimulate pancreatic secretion (115) perhaps by release of endogenous acetylcholine, as in the cat (116).

No reports are available about pancreatic exocrine function in patients with phaeochromocytoma. One patient with phaeochromocytoma developed recurrent pancreatitis, but the patient also suffered from stones in the bile duct (117).

Oestrogens

Effects of Oestrogens on the Exocrine Pancreas

The exocrine pancreas of both experimental animals and man has oestrogen receptors (118) probably on the acinar cells. However, there has been little direct study of the effects of oestrogens on the exocrine pancreas, although it appears that 17-beta-oestradiol inhibits basal and secretin-stimulated pancreatic secretion, especially of bicarbonate and water (119). These studies may be relevant to the oft reported association between use of oestrogen-containing contraceptive pills and the development of acute pancreatitis (120-126). It seems that in many patients, administration of oestrogens makes clinically evident a previously masked tendency to hyperlipidaemia, which then results in acute pancreatitis (120, 121), a course of events that has also been observed in postmenopausal women and men after prostatectomy who have been given oestrogen therapy (126). It was therefore recommended that oestrogens should not be given to women with type 4 hyperlipoproteinaemia. However, acute pancreatitis may develop in women who are not hyperlipidaemic (124) and may even be fatal (125). The mechanism by which oestrogens, or hyperlipidaemia, cause pancreatitis is not known, although pancreatic infarction has been proposed on the basis of one study (123). The effects of contraceptive pills on human pancreatic exocrine function have not been defined.

No reports of pancreatic exocrine function in patients with spontaneously excessive or defective

ovarian function are available. Relapse of pancreatitis with menstruation is apparently very unusual (127, 128).

While there is no record of systematic study of the effect of pregnancy on human pancreatic function, an association between acute pancreatitis and pregnancy has been recognized for many years (129). Experimental investigations in dogs showed a significant increase of all the indices of basal pancreatic secretion and there was an enhanced enzyme-secretory response to secretin and CCK during pregnancy, although secretory rate and bicarbonate output were unchanged (130, 131). The mechanisms by which the complex endocrine changes during pregnancy produce alterations in pancreatic exocrine secretion are not yet known, nor have any connections between these changes during acute experiments and the occurrence of pancreatitis been established.

Insulin

Effects of Insulin on the Exocrine Pancreas

It has long been suggested that the close anatomic relationship between the exocrine and endocrine pancreas reflects functional interrelationships between pancreatic islets and acinar tissue (132-136). The islet-acinar portal system of veins (137) results in high concentrations of insulin and other islet hormones reaching particularly the acinar tissue surrounding the islets. As a consequence, insulin has been considered to exert a trophic effect on the periinsular acinar cells, perhaps by stimulating the uptake of amino acids (138, 139). In autoradiographs of mouse exocrine pancreas, increased synthesis of amylase and a greater incorporation of labelled amino acids by the periinsular acini has been demonstrated, compared with other acinar tissue (140, 141).

At the physiologic level, insulin appears to augment the pancreatic exocrine response to stimulants such as CCK in animals (142). Sustained hyperinsulinaemia (in rats) increased the amylase content and total protein synthesis of the pancreas (143).

Insulin potentiates the release of amylase from the isolated rat pancreas and potentiates CCK-induced hyperpolarization of the acinar cells (144). Isolated mouse and rat pancreatic acinar cells were shown to specifically bind insulin, thus demonstrating the presence of insulin receptors (144a). The isolated acini also increased uptake of an incorporation of amino acids on exposure to insulin (145).

No reports of pancreatic exocrine function in patients with insulinomas are available.

The relationship between diabetes mellitus and exocrine pancreatic function has received quite a lot of comment, although much is inconclusive and contradictory. Several studies have shown that diabetic patients who have required insulin for more than five years, have grossly impaired pancreatic exocrine function (146–148) and often have histologically demonstrable lesions of the acinar, as well as islet cell, parts of the pancreas. These abnormalities are correlated with the duration of the disease, but not with its severity nor with the age of the patient (148). The causes of the pancreatic exocrine hypofunction have not been defined, but histologic studies of diabetic patients show acinar sclerosis and fatty change, resulting in some cases in almost complete disappearance of the acinar tissue (149). Similar appearances have been described in experimental animals with diabetes mellitus (150). Experimental diabetes has also produced functional changes in the exocrine pancreas. For example, experimental diabetes resulted in the disappearance of amylase from the exocrine pancreas and a decreased rate of protein synthesis (143). Other studies have confirmed that experimental diabetes reduces amylase synthesis, secretion and pancreatic amylase content, changes that are reversed by giving insulin (138, 151, 102). Biochemical studies have also shown that insulin restores to normal the incorporation of radioactively-labelled amino acids into amylase in rats made experimentally diabetic (140).

Effects of Pancreatic Disease on Insulin Secretion

Both excessive and defective islet cell function have been described in patients with acute and chronic pancreatitis and with pancreatic cancer, with increased or impaired secretion of insulin. The topic has been discussed previously (152).

Glucagon

Effects of Glucagon on the Exocrine Pancreas

Early reports demonstrated that in rat and rabbit pancreas, repeated injections of glucagon caused histologic alterations, mainly consisting of progressive degranulation and atrophy of the acinar cells (153–155). Subsequently, the effects of glucagon on the pancreas (156) were studied in the basal state and during hormonal stimulation of the pancreas, both in the dog (157–159) and in man (160). In the dog, glucagon did not affect basal pancreatic secretion, while administration of glucagon during stimulation with secretin and CCK produced a marked,

but evanescent decrease of the rate of secretion, bicarbonate output and especially enzyme output. Glucagon also reduced the bicarbonate-secretory response to duodenal acidification (158) and to food (161) in dogs. In man, glucagon administered by rapid intravenous injection in increasing doses significantly reduced the rate of pancreatic secretion and the concentration of protein (160). The pancreatic inhibition was dose-dependent. While the concentration of protein was reduced, no change was seen in the concentrations of bicarbonate or chloride, although bicarbonate output was, of course, reduced. The inhibitory effect occurred within five to ten minutes of the injection of glucagon and was followed by rebound hypersecretion. Since very marked suppression of enzyme secretion was reported with very small doses of glucagon, the possibility of physiologic rather than pharmacologic inhibitory effect for the hormone was inferred. Continuous intravenous infusion of glucagon also inhibited the pancreatic enzyme-secretory response to secretin and CCK in normal individuals and patients with acute or chronic pancreatitis. The glucagon had no effect on the pancreatic secretory rate or bicarbonate output (162).

The mechanism by which glucagon influences pancreatic exocrine secretion remains obscure. The hypothesis that the structurally related secretin and glucagon molecules (163) compete for receptor sites has not been supported by the most recent studies which have shown that glucagon does not exert inhibitory effects on secretin binding sites on pancreatic acinar cells (164) and have failed to show inhibitory interaction between secretin and glucagon. Moreover, unlike secretin and VIP, glucagon does not increase the intracellular content of cyclic AMP (164, 165). It seems probable that glucagon increases, rather than reduces, pancreatic blood flow (166), so that the secretory inhibition cannot be the result of reduction in blood supply to the gland.

In contrast with the above studies, glucagon given to feeding rats actually increased the pancreatic secretory rate and output of bicarbonate (167). Glucagon also increased the secretion of a blood-perfused isolated canine pancreas, by what was considered to represent a direct effect on the exocrine cells (168). Similarly, glucagon increased enzyme secretion from isolated pancreatic acinar cells (169), findings that were considered compatible with the observed degranulation of acinar cells and contrary to the inhibitory effects reported in the intact animals.

Perhaps some of the difficulties encountered in attempting to interpret the effects of glucagon on

the exocrine pancreas stem from the nature of the so-called inhibition. The difficulties in interpreting the significance of inhibition, followed by rebound hypersecretion in terms of overall positive or negative functions effects have been discussed previously (170). The effect of glucagon on pancreatic exocrine secretion is, under some circumstances, an example of just such inhibition-rebound phenomenon and may, therefore, be artifactual and of no functional significance.

Hyperglucagonaemia

While glucagon-secreting neoplasms have been described, no information is available about pancreatic exocrine secretion in glucagonomas.

Effect of Pancreatic Disease on Glucagon Secretion

Abnormal blood levels of glucagon have been reported in patients with the different types of pancreatic disease. In acute pancreatitis, hyperglucagonaemia has been noted to occur quite frequently (37, 58, 171, 172). However, other reports have denied that blood levels of glucagon are greater than normal in acute pancreatitis (34, 37) and, indeed, deficient alpha cell function, with secretion of abnormally small amounts of glucagon, has been considered a bad prognostic sign in acute pancreatitis (173).

In chronic pancreatitis, both hypo- and hyperfunction of the alpha cells have been recorded. The impaired secretion of glucagon tends to give rise to spontaneous hypoglycaemia, or excessive sensitivity to insulin hypoglycaemia, while excessive glucagon secretion contributes to the diabetic tendency observed in these patients (152).

Conclusions

We hope that the above accounts have emphasised the paucity and defective nature of our information about the function of the exocrine pancreas in endocrinopathic states. The lack of information is surprising, not only because many of the diseases are common, but also because the exocrine pancreas presents a unique opportunity of studying the effect of hormonal action on the synthetic and secretory processes of a gland engaged in producing exportable protein. Perhaps this report will stimulate some proper, systematic studies both of these processes in man, and of the interaction between the exocrine pancreas and diseases of the endocrine glands.

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Mycotic Pseudoaneurysm of the Left Ventricle: Report of a Case Successfully Treated

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Abstract

A retrocardiac mass was discovered in an 11-year-old girl during the course of treatment of femoral osteomyelitis. This apparently developed after a prolonged course complicated by *staphylococcus aureus* septicemia and a transient cardiac murmur. After extensive diagnostic examinations, exploratory thoracotomy revealed a sac-like structure consistent with pseudoaneurysm of the left ventricle. This was later excised under total cardiopulmonary bypass. The patient recovered uneventfully.

Aneurysm-like congenital and acquired abnormalities of the left ventricle in children are extremely rare. However, they can be life-threatening (1-7) and must always be considered in the differential diagnosis of a paracardiac mass.

A case successfully treated is presented. The diagnosis was not suspected preoperatively, but was made at exploratory thoracotomy.

Case Report

An 11-year-old girl was referred for management of right femoral osteomyelitis. Five months before admission, she developed swelling of the right knee following local trauma. Severe leg pain, fever, and malaise prompted prior hospital admission in another institution in which she was treated with parenteral antibiotics for six weeks. A heart murmur was noted during the course of treatment. An abscess localized within the swollen leg necessitated incision and drainage. She then developed a pathologic fracture of the femur that required applica-

tion of a hip spica. Five weeks later, the cast was removed and a persistent draining sinus was noted in the lower third of the thigh. Radiologic studies revealed chronic femoral osteomyelitis. At that point, the patient was transferred to The Mount Sinai Hospital.

Physical examination was entirely unremarkable except for a draining sinus on the right thigh. Pertinent laboratory data included: hemoglobin 12.3 gm%, hematocrit 38%, white blood cell count 6,900 cells/mm³ with 28% segmented cells, 66% lymphocytes, 3% monocytes, and 3% eosinophils. All serum chemistries were within normal limits except for mild elevations of alkaline phosphatase and lactic dehydrogenase. The initial electrocardiogram demonstrated T-wave inversion in the anterior precordial leads but subsequent tracings were normal. Radiograms and tomography of the thorax revealed a large solid mass in the left lower lung field posterior to the cardiac shadow (Fig. 1).

A biopsy of the femur demonstrated osteomyelitis. A sequestrectomy and curettage of the femur was subsequently performed. Culture of the sequestrum grew *staphylococcus aureus*. The patient became febrile on the night of surgery but defervesced upon therapy with parenteral oxacillin and, later, with oral dicloxacillin.

Additional noninvasive examinations of the chest lesion were nondiagnostic. Aortography did not demonstrate pulmonary sequestration.

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FIG. 1. Chest radiogram obtained on admission, showing a round radiodense mass superimposed with the cardiac shadow at the apical area.

Thereafter, exploration through a left posterolateral thoracotomy was performed. There was a 4×5 cm intrapericardial mass that appeared to be thin-walled, contiguous with the posterior wall of the left ventricle and adherent to the overlying parietal pericardium. The sac was pulsatile and could be emptied on manual compression. Excision was not attempted and the chest was closed. After recovery from surgery, left ventricular cineangiography was performed and revealed a small stream of dye that seemed to exit through the posterior wall of the left ventricle and subsequently disappeared without opacifying the mass. The findings at the previous thoracotomy strongly suggested a left ventricular diverticulum or aneurysm and open intracardiac operation was advised.

Under total cardiopulmonary bypass with profound hypothermia and cardioplegic arrest, a thin, fibrotic 4 × 5 cm diverticulum was found to com-

municate with the left ventricular cavity through two 3 mm openings in the posterior wall (Figs. 2, 3). The fistulous tracts were surrounded by dense fibrous tissue suggestive of a prior inflammatory process. The sac was excised and the fistulous openings sutured closed.

The postoperative course was uneventful. When seen eight months postoperatively, the patient was afebrile and completely asymptomatic. Histologic examination of the sac demonstrated hyalinized collagenous tissue. No myocardial fibers were identified. Intraoperative cultures of the specimen were sterile.

Discussion

Localized sac-like structural abnormalities of the left ventricle in children can be of either congenital



FIG. 2. The diverticulum on the posterior wall of the left ventricle is shown in situ. The heart is pulled anteriorly with apical traction sutures and the diverticulum has been opened. The edges of the thin wall are retracted to expose the smooth inner surface. A fine suction tip has been inserted into one of the fistulous openings.

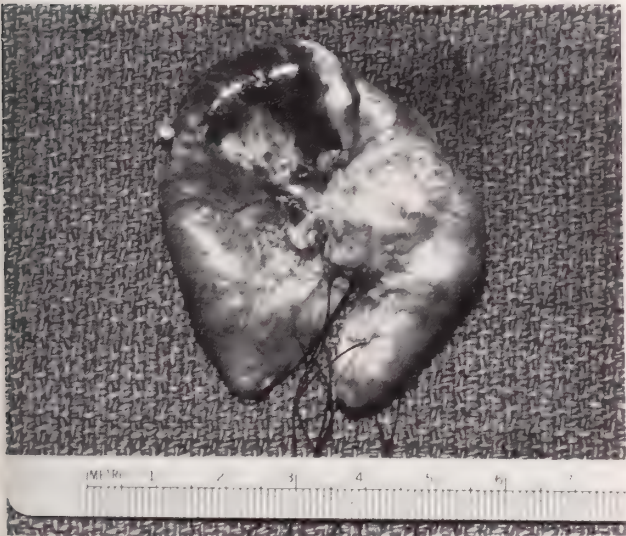


FIG. 3. The excised diverticulum showing dimensions.

or acquired etiology. A number of terms have been employed to characterize these lesions (aneurysm, diverticulum, pseudoaneurysm). As observed by Treistman et al (8), these terms unfortunately have been used interchangeably in the literature. Al-

though both aneurysms and diverticula can be of congenital etiology, the term “pseudoaneurysm” has been consistently used to describe a lesion of acquired origin. Development of a pseudoaneurysm implies a relatively controlled rupture. Hurst (9) points out that the development of a false aneurysm requires time for formation of fibrous thickening sufficient to contain the escaping blood within a restricted area. The site of rupture which becomes the neck of the pseudoaneurysm is, therefore, usually small.

Both congenital left ventricular diverticula and acquired pseudoaneurysms may appear as narrow-necked sacs. The presence of endocardium or myocardial fibers on microscopic examination of the sac wall would largely rule out pseudoaneurysm but their absence is not conclusive (8). Correlation with the clinical picture, more than gross or microscopic pathology, may therefore prove to be a more decisive factor in establishing the final diagnosis of pseudoaneurysm.

In this case, the clinical association of the pseudoaneurysm with presumed seeding from the staphylococcal bone infection is significant. The finding

of a new cardiac murmur during the initial episode of sepsis is suggestive of a concomitant infectious process involving the heart. The mechanism of pseudoaneurysm formation here is probably similar to that previously described by Pirani (6) and Weiss (7) who observed that myocardial perforation resulted from abscess formation either secondary to septic emboli or as a result of endocarditis with extramural extension. The latter mechanism is invoked in pseudoaneurysms resulting from perforation of the mitral-aortic intervalvular fibrosa (4, 5). In our case, the former mechanism seems more probable since the perforation was located in the posterior wall of the left ventricle.

A similar case has been reported by Beare (10). The patient succumbed from complications following a technically successful repair. Indeed, upon search of the English literature, all reported mycotic pseudoaneurysms except for two other cases by Yakierevitch, et al (11), have ended in the patient's early demise. The present patient's survival is encouraging; the outlook of these problems may not be as dismal as earlier experiences would have suggested.

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Anesthetic Management of Children for Cardiovascular Investigation

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In recent years, major advances in surgical correction of congenital heart abnormalities have necessitated ever increasing accuracy in the diagnosis of these abnormalities. Such accuracy demands that clinical findings be complemented by cardiac catheterization and angiocardiology. In most medical centers in which children with congenital heart disease are investigated, the anesthesiologist is a member of the cardiac diagnostic team.

The management of the child undergoing cardiac catheterization requires close cooperation between the cardiologist and the anesthesiologist. These patients are often critically ill and present special problems to the anesthesiologist who must be familiar not only with the problems of pediatric anesthesia but also with the pathophysiology of congenital heart disease and the risks involved in cardiac catheterization. The anesthesiologist who is aware of the complications that occur in such patients during cardiac investigation is able to prepare, in advance, for corrective measures that may be lifesaving (1-3).

The purpose of this paper is to present the management of infants and children undergoing cardiac catheterization, and to discuss the most common problems and complications that may occur during the procedure.

General Considerations and Risks

Cardiac catheterization is now an accepted method of investigation in children with congenital heart disease. It must be realized, however, that it is a major procedure that may terminate in the death of the patient. It is difficult to quote statistical

mortality figures that are valid. Braunwald and Swan (4) in their combined study published in 1968 showed a major complication rate of 3.6%. During angiocardiology, the complication rate was 4.5%. There was an overall mortality of 0.45%, but this rose to 6% in infants of less than two months of age. Recent statistics have shown that mortality following cardiac catheterization in infants has now been reduced to less than 1% (5). However, when the investigation is necessary in the first month of life the inhospital mortality is still considerably greater than 1%. The hazard of catheterization cannot be clearly separated from those of the cardiac lesion or its treatment.

Cooley et al (6), have stated that over 80% of infants born with congenital heart disease are suitable for definitive or palliative surgical correction provided that the diagnosis is made early. As the emphasis of treatment of congenital heart disease has shifted towards the first few days and weeks of life, an increasing number of cardiac catheterizations have been performed in the newborn and young infant (7, 8). Most of these infants are seriously ill. They have complicated congenital heart disease and cardiac catheterization is performed as an emergency procedure. Major problems encountered are severe congestive heart failure, cyanosis, and hypoxia. Many of these patients are prone to develop serious complications during cardiac catheterization and require constant attentive monitoring by the anesthesiologist during the procedure. Following the period of infancy, cardiac catheterization studies are usually performed on an elective basis in children who have, in most instances, a single cardiac abnormality (interventricular septal defect or arterial septal defect, etc.) with relatively low risk. These patients are not as symptomatic as the previous group, but almost all require heavy sedation or general anesthesia during cardiac catheterization.

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The information that cardiac catheterization provides includes pressures and oxygen content of the blood in the various chambers of the heart and the presence of abnormal communications between these chambers. Generally, blood sampling and pressure recordings are not carried out simultaneously but are measured consecutively. A steady cardiopulmonary state is, therefore, essential during the investigation to allow comparison of pressures with oxygen saturations observed at any one stage of the procedure. The role of the anesthesiologist is directed towards the care and comfort of the patient and the provision of a stable cardiovascular system and metabolic rate (10).

Anesthetic Techniques

There is a wide choice of methods for the management of patients undergoing cardiac catheterization. This variety indicates that no one technique is entirely satisfactory (10).

Anesthesia must provide conditions in which the patient is able to remain at rest for a prolonged period. There must also be no significant disturbance of hemodynamics and no interference with the analysis of blood gases. These conditions must be provided in the darkness of an x-ray room in a patient who may be a very poor risk. The most commonly used techniques have included rectal thiopental (9, 10), the "lytic cocktail" (chlorpromazine or promazine, promethazine, and meperidine) (11-14), diazepam (15), ketamine (17-20), Innovar® (droperidol and fentanyl) (16), and general anesthesia (9, 10, 21).

In many centers, infants under six months of age are catheterized without premedication or anesthesia. All procedures are performed under local anesthesia. The dose of local anesthesia used for the cut down site is critical in these small infants. Lidocaine 3mg/kg without epinephrine is the appropriate maximum dose. Particular care must be exercised in these patients with high pulmonary vascular resistance or in severely cyanotic infants, that hypoventilation is prevented. It is probably wiser to avoid sedation in critically ill infants. General anesthesia with endotracheal intubation to allow adequate ventilation may be indicated in such cases.

In children over the age of six months, catheterization is usually possible after the administration of suitable sedatives. This may be a barbiturate, an opiate, or a combination of drugs. In those children who are thought unlikely to be easily managed by simple sedation, one of the following techniques are commonly employed:

Sedative Techniques

The "lytic cocktail" (11-13), described by Smith, Rowe and Vlad is a technique in which they gave a mixture containing chlorpromazine 6.25 mg, promethazine 6.25 mg, and meperidine 25 mg in one milliliter of the mixture. It was given intramuscularly one hour before transfer to the catheterization room. The dose varied according to the age and condition of the patient. Noncyanotic infants and children received 1 ml/10 kg up to a maximum of 1.5 ml. Deeply cyanotic infants were given half this dose. The dose may be increased if necessary at the time of catheterization by injection of a dilute solution through the catheter. More recently, Ordish and Mair (12, 13) have replaced chlorpromazine with promazine. In general this combination of drugs produced adequate sedation; however, there have been reports describing disadvantages of this technique. Smith et al (11), reported that some children were not adequately sedated during the investigation despite additional intravenous supplementation. When large supplementary doses of the mixture were used, recovery after catheterization was prolonged. The mixture, given intravenously, was not conducive to the maintenance of a steady cardiovascular state. Even when chlorpromazine was omitted, the mixture was found to produce significant hypotension and tachycardia (13, 14). Healy, has described the use of diazepam as an intravenous supplement to the above premedication in doses of 1 mg/kg (15). The majority of his patients showed no alteration in blood pressure (73%) or heart rate (76%).

MacDonald et al (16), have described the use of neuroleptic analgesia for diagnostic cardiac catheterization studies. They reported falls in pulmonary vascular pressure particularly when droperidol (or phenoperidine) was used and this may cause errors in assessment.

In recent years, ketamine anesthesia has achieved widespread popularity for its unique property of inducing analgesia and anesthesia without circulatory depression (17-20). Following atropine premedication, an initial dose of 5-10 mg/kg is given intramuscularly with supplementary doses of 1 mg/kg/IV when necessary. The initial injection is followed by an increase in cardiac output, heart rate, and systemic arterial blood pressure. The pulmonary artery pressure also rises, but this probably reflects an increase in cardiac output rather than a direct effect on the pulmonary vascular resistance. These hemodynamic effects are transitory and are barely detectable after 15 minutes. Insignificant changes follow subsequent injections. Many au-

thors have reported these cardiovascular effects may be minimized by slow intravenous injection. Ketamine should, however, probably not be used in coarctation of the aorta associated with hypertension or severe left ventricular obstruction. In our institution in the past five years, more than four hundred children underwent cardiac catheterization under ketamine anesthesia. The incidence of undesirable cardiovascular disturbance or postoperative restlessness and hallucinations was negligible.

General Anesthesia Techniques

General anesthesia permits greater flexibility in the management of the child undergoing cardiac catheterization. However, since the introduction of the "lytic cocktail" and ketamine, the need for general anesthesia during cardiac catheterization has become infrequent. General anesthesia may be used as a supplement to heavy sedation, or may be undertaken from the onset for those very ill patients when intubation and oxygenation are considered to be necessary before the investigation can proceed (9, 10).

Many anesthetic agents have been used in the past for general anesthesia, including intravenous barbiturates, divinyl ether, diethyl ether, trichlorethylene, halothane and enflurane (9, 10, 21–26). Sedatives are usually given for premedication in children over six months of age. Atropine provides an effective decrease in vagal reflexes. In young infants, intubation is generally performed awake after preoxygenation. In older children, anesthesia is induced with an intravenous barbiturate or by inhalation of nitrous oxide with oxygen and a low concentration of halothane or enflurane. Endotracheal intubation is carried out with the aid of a muscle relaxant. Dysrhythmia, especially bradycardia, after the initial dose of succinylcholine is very common in children. Atropine 0.01 mg/kg/IV given just prior to the succinylcholine decreases the incidence of dysrhythmia. Following intubation, anesthesia is continued with 30%–50% oxygen in nitrous oxide, with or without additional halothane or enflurane, using assisting or controlled ventilation. Minor deviations of cardiopulmonary performance produced by IPPV are more than compensated for by avoidance of agents which affect myocardial contractility, peripheral resistance and development of dysrhythmias. Higher concentrations of oxygen (100%) are used only if there is definitive pulmonary pathology, or if the patient is critically ill. In general, halothane is avoided in infants with

left ventricular obstruction or severe congestive heart failure (27).

Complications

All intracardiac procedures are potentially dangerous. Even in the best hands there is always a risk to life (2, 3, 8). The cardiac and pulmonary function must be monitored constantly and vigilantly. The cardiovascular system in patients with congenital heart disease is capable of rapid alterations. Hypoxia, for example, resulting from even a minor airway obstruction may be disastrous. In almost all cases, use of the team approach makes early detection of the complication possible, with minimum risk to the patient. In general, complications during cardiac catheterization involve the cardiovascular and pulmonary systems and may be related to catheter manipulation or to anesthetic medications. Other complications occur secondarily to exposure or to metabolic derangement or may occur following angiocardiographic studies (28).

Cardiovascular Complications

Cardiac Dysrhythmias

Even with the most skillful manipulation of the catheter, dysrhythmias are unavoidable during cardiac catheterization. Immediate removal of the catheter from the dysrhythmia-producing focus usually allows the rhythm to return to normal. The likelihood of dysrhythmia is also reduced by ensuring adequate oxygenation and controlling blood carbon dioxide levels (28, 29).

Tachydysrhythmias

Supraventricular tachycardia is the most common. Persistent tachycardia observed during cardiac catheterization frequently can be converted to a slower rhythm by manipulation of the catheter. If this is not successful, edrophonium 0.2 mg/kg/dose IV may stop the dysrhythmia. Rapid digitalization may also be indicated. Sustained hypotension with persistent supraventricular tachycardia requires immediate direct current defibrillation. During this period of dysrhythmia, adequate ventilation must be maintained, if necessary, by positive pressure ventilation.

Ventricular Arrhythmia

Persistent ventricular premature beats or bigeminy often can be abolished by intravenous administration of a single dose of lidocaine 1 mg/kg/IV or

diphenylhydantoin 200 mg/kg/5 min intravenously. If ventricular tachycardia or fibrillation occurs, immediate direct current defibrillation must be performed while the circulation is maintained by closed chest massage and adequate ventilation (1, 4, 5).

Bradycarrhythmias

The most common bradycardias seen during cardiac catheterization are sinus bradycardia and heart block.

Sinus bradycardia is particularly dangerous in infants if it is associated with hypotension, and respiratory distress. Unexplained bradycardia should alert the anesthesiologist to the possibility of hypoxia and this should be excluded before resorting to drugs for symptomatic treatment. Atropine in this situation may have only a temporary effect.

Complete heart block is an unusual complication, but it may occur during manipulation of the catheter in the region of the tricuspid valve. Infusion of Isoproterenol 0.5-2 mg in 100 D5W or 0.0-0.5 micrograms/min IV is indicated if the block persists. If this fails, transvenous pacing should be initiated.

Hypoxia

In patients with congenital heart disease marked hypoxemia is usually a result of intracardiac right-to-left shunting. Arterial hypotension secondary to sedation or dysrhythmia may increase the right-to-left shunt. Hypoxic spells may occur during investigation and require immediate treatment with oxygen, morphine, sodium bicarbonate, and propranolol (8, 29).

Cardiac Tamponade

The myocardium may be punctured while the catheter is being manipulated or during angiocardiology following injection of dye. Cardiac tamponade could prove to be rapidly fatal, especially if the atrial/appendage has been perforated. In such cases the catheter should be allowed to remain in situ and immediate surgery is indicated (31).

Overloading of the circulation with fluid used in flushing the catheter system must be avoided particularly in patients with congestive heart failure and in small infants. In cyanotic patients, on the other hand, adequate amounts of intravenous fluid must be given to prevent dehydration. In cyanotic patients, oral fluids should not be withheld for a long period without adequate intravenous fluid replacement.

Disturbance of Blood Volume

Considerable blood loss can occur during cardiac catheterization particularly during arterial cannulation following multiple blood sampling in the small infant. Acute anemia secondary to blood loss may cause congestive heart failure in patients with large shunts or hypoxic spells in infants with a tetralogy of Fallot. Blood should be replaced.

Complications Related to Medications and Anesthesia

Respiratory Depression

Medications given for sedation may cause respiratory depression often as a result of an inappropriate dose or after supplementation during the procedure. Small and cyanotic infants are particularly sensitive to morphine or meperidine. If respiratory depression is accompanied by a bradycardia and fall in blood pressure, the possibility of hypothermia or acidemia should also be considered. Adequate ventilation must be ensured during cardiac catheterization. If this cannot be maintained, positive pressure ventilation must be used.

Convulsions

Generalized seizures may occur if large doses of lidocaine are used for local anesthesia or if accidental intravascular injections occur. Generalized convulsions have also been described in children with a history of epilepsy who were anesthetized with ketamine (32).

Complications Related to Exposure and Acid-base Disturbance

Hypothermia must be avoided particularly in infants and small children. Adverse effects include hypoventilation, metabolic acidosis and cardiac dysrhythmias. A minimal rectal temperature of 96° F should be maintained in small infants by placing them on a warming mattress. A heating lamp may be of greater value in this situation. The most important factor in temperature maintenance in the cardiac catheterization laboratory is room temperature which should be set at 80° F for newborns and 78° F for children of 2-3 months of age (33).

Acid-base Disturbance

A metabolic acidosis is the predominant type of acid-base disturbance found before and during car-

diac catheterization studies, particularly in cyanotic infants. This should be promptly corrected with sodium bicarbonate (34).

Hypoglycemia

It has been well demonstrated that infants with severe hypoxemia or with peripheral circulatory failure frequently develop severe hypoglycemia. In these infants a blood glucose level should be measured routinely. Blood sugar may be roughly estimated by the Dextrostix reagent strip. If the level is considered to be low, 10% dextrose should be administered promptly (5, 28).

Complications Related to Angiocardiographic Studies

Since cardiac catheterization is inseparable from the angiocardiographic aspect, it is important to consider the influence that the introduction of contrast material might have in the morbidity or mortality. The toxic effects of the contrast material would appear to be caused partly from the medium itself and partly from the hemodynamic alteration induced (35). Standen (36) has stated that injection of a contrast material results in increase in the serum osmolality. This, in time, causes extravascular fluid to be drawn rapidly into the vascular compartment increasing the plasma volume and decreasing hematocrit values. The increase in plasma volume could be deleterious to patients in heart failure. The neurologic complication of convulsions induced by hypertonic effects of angiocardigraphy contrast material can be avoided by the spacing of injections and careful avoidance of excessive amount of contrast material. As contrast material is added, it may cause metabolic acidosis in large doses especially in small infants. It is important to correct acidosis before proceeding to angiocardiography.

Conclusions

Cardiac catheterization should not be denied to any patient with congenital heart disease. In 1955, Sones stated (37) that "no patient should be denied the opportunity for precise diagnosis and possible cure or improvement because he is too small or because functional severity of the lesion appears so great that death might occur during or after catheterization. For such a patient the greatest hazard lies in continuing failure to make an exact diagnosis upon which effective treatment may be based."

Since an improved survival rate in congenital heart disease depends primarily now on increased salvage in the first year of life, and since a definitive diagnosis must precede any therapeutic measures, early investigation remains the first step in the management of these patients.

Knowledge of complications occurring commonly in the cardiac catheterization laboratory will lead to their prevention; thus if complications do occur, then treatment will be prompt, rational, and effective (38). The anesthesiologist, because of his daily experience in monitoring, evaluating, and treating cardiopulmonary alterations, has a unique opportunity and obligation to help the child with severe congenital heart disease undergo the stress of cardiac catheterization with relative safety and with fewer complications.

Summary

The management of the child undergoing cardiac investigation requires close cooperation between the anesthesiologist and the cardiologist.

The various techniques of anesthesia used for cardiac catheterization and angiocardiography have been reviewed. The risks of cardiac catheterization and complications have been presented with particular emphasis on their prevention and treatment.

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Successful Medical Treatment of a Tuberculosis B-E Fistula

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Abstract

An early tuberculous bronchial esophageal fistula was diagnosed in a patient with a myeloproliferative disorder being treated with corticosteroids. Although tuberculous bronchial esophageal fistulas have traditionally been treated by surgical means, our patient was successfully treated by conventional medical modalities including antituberculosis therapy, hyperalimentation, and discontinuation of the steroids.

We suggest that medical management of this rare complication of tuberculosis is appropriate and continue to be tried in the future.

It has long been recognized that the corticosteroid treatment may activate a latent tuberculosis infection (1). This problem is further amplified in patients afflicted with hematologic neoplasia, for not only may there be an increased susceptibility to mycobacterium infections in these patients (2), but also the protocols used for the treatment of these disorders may contain corticosteroids.

In the following case report, a latent tuberculosis infection activated by the use of prednisone in a patient with a myeloproliferative disorder resulted in the formation of a bronchial esophageal fistula. Fortunately, this complication was recognized early thus allowing successful medical management in this man who would have been a high-risk surgical patient.

Case Report

A 50-year-old man was referred to us because of fever and difficulty swallowing two weeks after the initiation of prednisone therapy for his myeloproliferative syndrome.

He had been well until two months previously when he noted easy fatigability. At one month prior to admission, he sought the services of a physician.

Initial physical examination was normal, except for the presence of pallor. The hemoglobin was 8 grams percent, the white count was 3,000/ul with 22 percent segs, 5 percent Pelger Huet cells, 55 percent lymphs, and 18 percent monocytoid cells. His platelet count was 40,000. The SMA 12 was normal. The serum protein electrophoresis was normal.

A marrow aspiration showed increased cellularity with a markedly increased M:E ratio of 20 to 1. Pelger Huet cells and other cells with myelodysplastic features were noted. The karyotype of the marrow cells was 46XY. No Philadelphia chromosome could be demonstrated.

The diagnosis of a myeloproliferative syndrome was made. Prednisone, 60 mg daily, was started. Ten days later, the patient complained of dysphagia, weight loss and fever.

For these reasons, he was transferred to the Green Hospital of Scripps Clinic.

The physical examination showed no abnormalities, except for an oral temperature of 102°F, pallor, and a spleen tip felt 2 cms below the left costal margin. Although he could swallow solid food with minimal difficulty, attempts at swallowing liquids resulted in expectoration of small quantities of the liquid.

Placement of an intermediate strength PPD resulted in 25 mm of induration. Although the roentgenograms of the chest were normal, an esophagram demonstrated a small fistulous tract connecting a secondary bronchus in the left upper lobe with the

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FIG. 1. Esophagram demonstrating a small, fistulous tract between a secondary bronchus in the left upper lobe and the esophagus.

esophagus (Fig. 1). The sputum demonstrated a small number of acid fast bacilli and subsequently grew mycobacterium tuberculosis sensitive to all common antituberculous medications.

The patient's prednisone was discontinued. He was started on isoniazid 300 mg, rifampin 600 mg, ethambutol 1400 mg and pyridoxine 50 mg, each given daily. He was kept NPO and hyperalimented over the next three weeks with intralipidTM, freeamineTM and dextrose. Seven days after the start of such therapy, he became afebrile. A repeat esophagram at three weeks was normal. Oral food was started and tolerated well. He was discharged from the hospital. By three months after discharge, he had gained 25 pounds in weight, continuing to have no difficulty in swallowing. He tolerated his antituberculosis drugs without ill effects. Unfortunately, his pancytopenia persisted and at six months after discharge he was noted to have blasts in his

blood, a platelet count of 5,000 and a marrow aspirate filled with myeloblasts. The patient subsequently died of a central nervous system bleeding during an attempt at inducing remission of his acute myelogenous leukemia. Permission for autopsy was not granted.

Discussion

Acquired fistulas between the tracheobronchial tree and the esophagus are a rare manifestation of mycobacterium infections. When last reviewed, only 22 cases had been reported in the world's literature (3).

Our patient, a man with a myeloproliferative disorder which later manifested itself as AML, had his latent tuberculous infection activated by the use of prednisone. That corticosteroids may activate latent tuberculous infections is well known. Also long recognized is a relationship between hematologic neoplasia and tuberculosis, with some investigators suggesting that the leukemoid reaction of tuberculosis may resemble the blood picture seen in acute myelogenous leukemia (AML) (4) while others stress that patients with hematologic neoplasia have an unusual susceptibility to developing tuberculosis infections (2).

That our patient had a tuberculous infection was initially suggested by his strongly reactive PPD. We wish to emphasize that all patients with hematologic disorders should have a PPD performed if high dose steroid treatment is contemplated.

Our case represents the second reported case of a tuberculous BE fistula that was successfully treated by medical intervention alone. As in the previously reported case (3), there was no obvious pulmonary infiltrate in our patient at any time, the fistulous tract was small, and the number of mycobacterium in the sputum was low. We suggest that when these circumstances are present, such fistulas can be successfully treated with medical therapy alone, utilizing antituberculous medications and total parental alimentation.

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Verruca Planavulgaris

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The histologic features of the common and plane warts are well defined in all textbooks of dermatopathology. During the past two years, I have noted and collected fifteen cases of a verrucous skin lesion that show histologic features of both but lack the total characteristics of each. They represented 4.5 percent of all verrucae examined during this period. I have chosen to call this lesion verruca planavulgaris which represents the subject of this report.

Materials and Methods

Fifteen lesions were biopsied from fifteen patients and were fixed in Bouin's solution, routinely processed, embedded in paraffin, sectioned, and stained with hematoxylin and eosin.

The clinical data are summarized in Table 1. The lesions ranged from skin-colored to tan, slightly elevated, smooth or verrucous firm papules that in size were from 0.2 to 0.8 cm. in maximum dimension. The majority of lesions involved the dorsa of the hands and wrists (six cases) and the face (four cases). Two lesions were located on the knees, one on the elbow, one in the axilla and one on the anterior thigh. Sex distribution was approximately equal. The base of each lesion was cauterized following biopsy and there have been no recurrences.

Examination of each case by light microscopy showed similar histologic features (Figs. 1-3). One observed slightly to moderately papillomatous lesions covered by an epidermis showing varying degrees of hyperkeratosis, parakeratosis, hypergranulosis and acanthosis associated with slight to marked elongation of rete ridges. Many of the cells within the upper malpighian and granular layers were enlarged with vacuolated cytoplasm and centrally located nuclei. The nuclei were either normal

in size, round, oval, or indented with a fine, pale chromatin network associated with small nucleoli, or shrunken, irregular and pyknotic. The thickened horny layer varied from the normal loose basket-weave appearance to markedly compact. Subepidermal vascular ectasia was absent in most cases, but was observed to a slight degree in some. No inclusion bodies, intranuclear edema or coarse basophilic or eosinophilic cytoplasmic granules were identified in any of the lesions.

TABLE I
Verruca Planavulgaris

Case	Age	Sex	Location	Size*	Duration	Clinical Impression
1	61	M	Elbow	0.5 cm	11 mo	Seborrheic keratosis
2	23	F	Jaw	0.2 cm	1 mo	Verruca plana
3	27	M	Knee	0.4 cm	5 mo	Verruca vulgaris
4	28	M	Dorsum of hand	0.3 cm	2 mo	Verruca vulgaris
5	26	M	Dorsum of hand	0.4 cm	3 mo	Verruca plana
6	30	F	Knee	0.6 cm	1 mo	Psoriasis
7	37	F	Cheek	0.3 cm	3 mo	Verruca plana
8	39	M	Forehead	0.7 cm	5 mo	Seborrheic keratosis
9	18	F	Cheek	0.3 cm	1 mo	Verruca plana
10	70	F	Dorsum of hand	0.5 cm	4 mo	Verruca plana
11	25	M	Dorsum of wrist	0.4 cm	6 mo	Seborrheic keratosis
12	55	M	Anterior thigh	0.8 cm	12 mo	Seborrheic keratosis
13	29	F	Dorsum of wrist	0.5 cm	7 mo	Verruca vulgaris
14	57	F	Dorsum of hand	0.6 cm	9 mo	Basal cell carcinoma
15	42	F	Axilla	0.4 cm	6 mo	Verruca plana

* Size represents maximum dimension.

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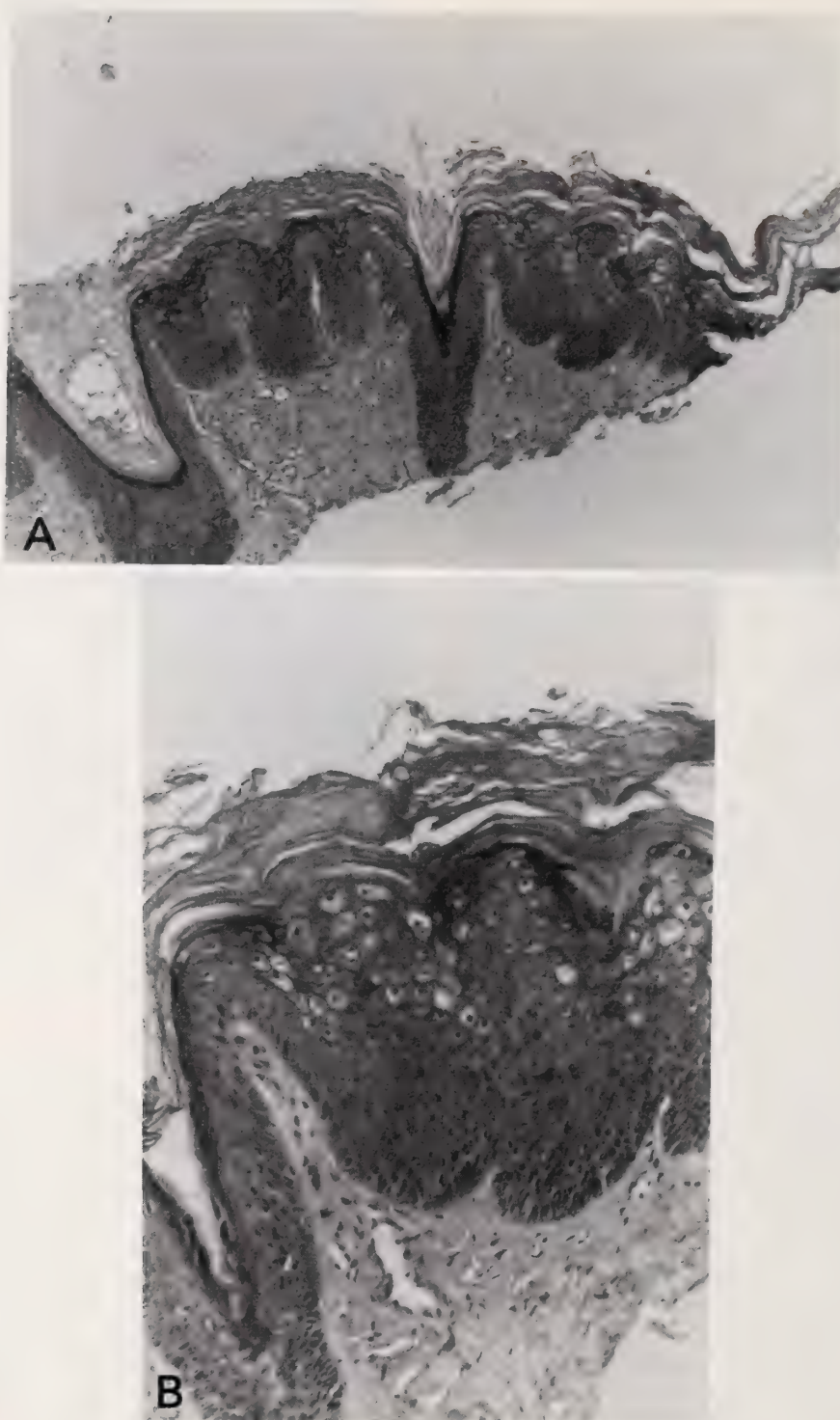


FIG. 1A. Scanning view of case 1 showing slightly papillomatous lesion associated with a moderately compact horny layer and a moderate degree of spotty hypergranulosis and parakeratosis. The characteristic cytoplasmic vacuolar changes are present within the upper malpighian and granular layers. Basketweaving and subepidermal vascular ectasia are slight (H & E $\times 40$).

FIG. 1B. Higher magnification of Fig. 1 A detailing features (H & E $\times 100$).

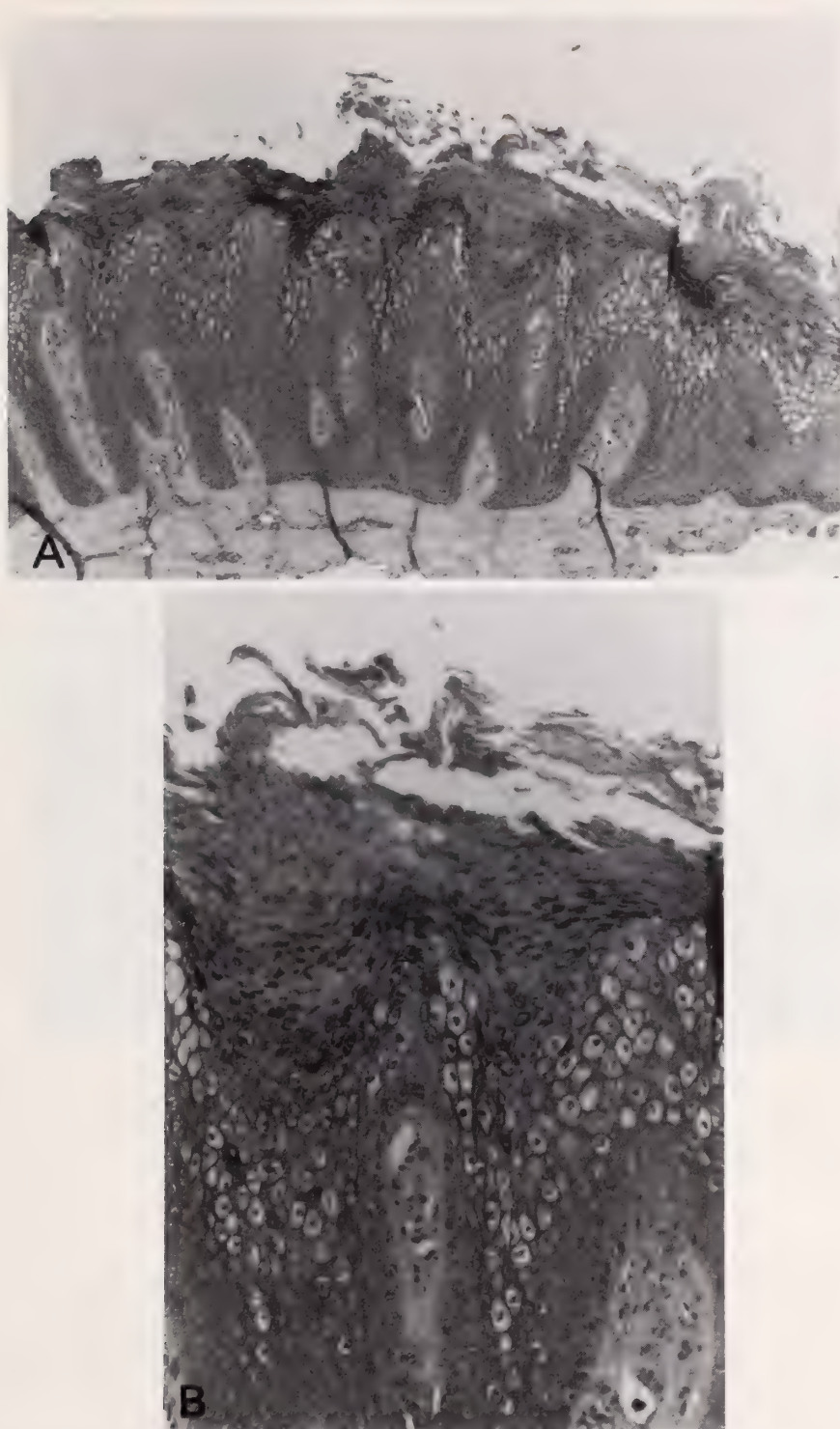


FIG. 2A. Scanning view of case 6 showing two moderately papillomatous lesions with features similar to case 1 except for a marked degree of compact hyperkeratosis and parakeratosis and an absence of basket-weaving (H & E $\times 40$).

FIG. 2B. Higher magnification of Fig. 2A detailing features (H & E $\times 100$).

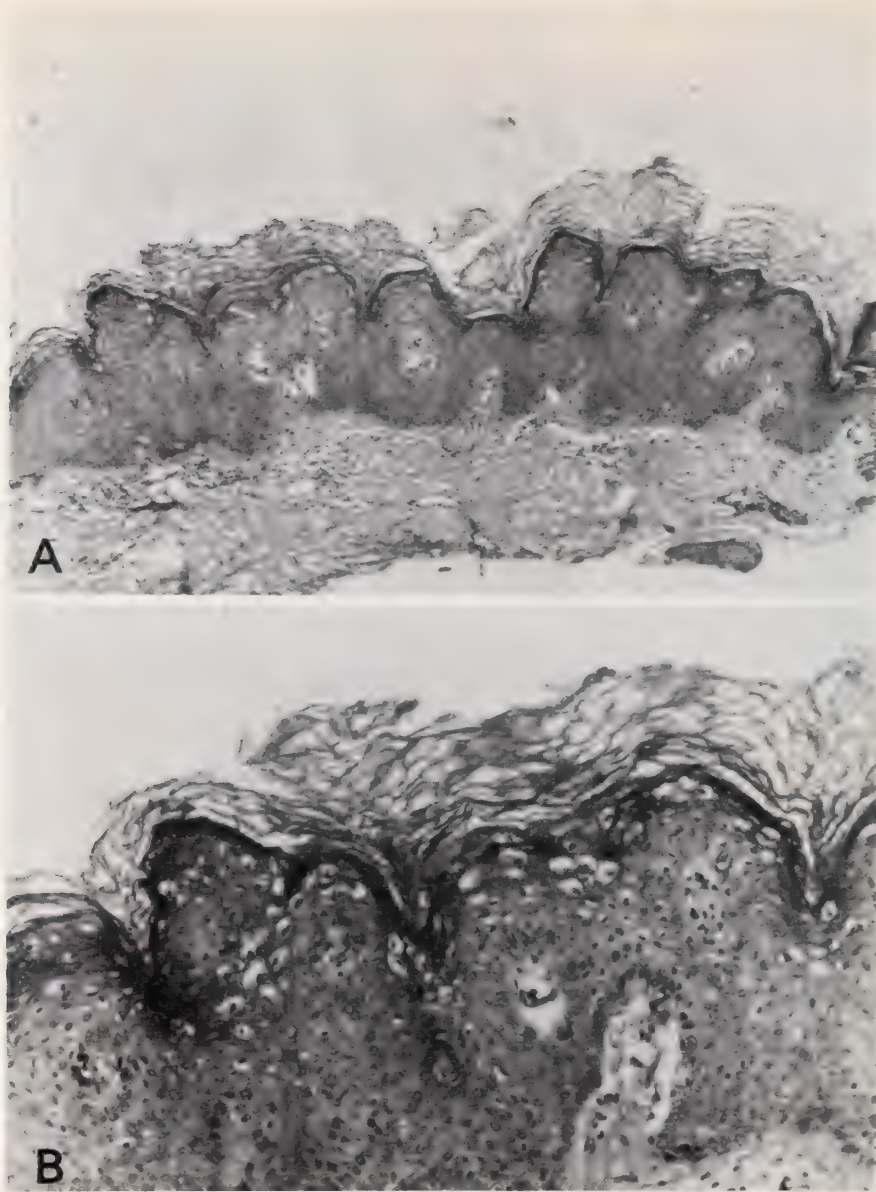


FIG. 3A. Scanning view of case 10 showing a markedly papillomatous lesion associated with a marked degree of basket-weaving and a slightly compact horny layer. Hypergranulosis, parakeratosis and subepidermal vascular ectasia are slight. The characteristic cytoplasmic vacuolar changes are present (H & E $\times 40$).

FIG. 3B. Higher magnification of Fig. 3A detailing features (H & E $\times 100$).

Discussion

While the clinical features of some cases were characteristic of a verruca plana, other cases were considered to represent verrucae vulgares. Microscopically, each lesion showed some histologic features of both. Table II gives a comparative summary of their histopathology. The hypergranulosis, nuclear findings and cytoplasmic vacuolar changes are characteristic of a verruca plana. In some cases the degree of acanthosis and elongation of rete

ridges was comparable to that observed in a verruca vulgaris. The papillomatosis, parakeratosis and compact hyperkeratosis are features observed in a verruca vulgaris. Other characteristics of the latter were lacking.

In a previous communication (1), I reported the possible emergence of a fourth histologic variant of wart, the signet ring verruca. The verruca plana-vulgaris seems to represent yet a fifth histologic variant of wart, probably caused by the same virus. It is tempting to speculate that this variant repre-

TABLE II
Comparative Histopathology with Light Microscopy

Histopathology	Verruca Plana	Verruca Vulgaris	Verruca Planavulgaris
Papillomatosis	—*	++ to +++	+ to ++
Compact hyperkeratosis	—	++ to +++	+ to +++
Loose basket-weave hyperkeratosis	++ to +++	—	— to +++
Parakeratosis	—	+ to +++	+ to +++
Hypergranulosis	+ to ++	+ to ++	+ to ++
Acanthosis	++	++ to +++	++ to +++
Elongation of rete ridges	— to +	++ to +++	+ to +++
Enlarged cells with vacuolated cytoplasm and central shrunken or normal-sized nuclei in granular and horny layers	+++	—	+++
Subepidermal vascular ectasia	—	+ to +++	— to +
Enlarged vacuolated nucleus in granular layer	—	Often present	—
Intranuclear inclusion bodies in granular and horny layers	—	Often present	—
Coarse basophilic or eosinophilic granules in granular layer	—	Often present	—

* — = not observed
+ = slight
++ = moderate
+++ = marked

sents a transition stage in the transformation of a plane to a common wart. It is also possible, in the light of recent evidence (2, 3), that this histologic variant of wart might be caused by a specific anti-

genic type of human papilloma virus. I hope to confirm the viral etiology of this lesion by electron microscopy in the near future.

Summary

Fifteen verrucae showing histologic features of both the plane and common wart were observed during the past two years, representing 4.5% of all warts examined during that period. It is therefore suggested that it be called verruca planavulgaris. It seems to represent a fifth histologic variant of wart and might possibly be either a transition stage in the transformation of a plane to a common wart or a wart produced by a specific antigenic type of human papilloma virus.

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Fibrous Histiocytoma of the Parotid Gland

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Nonepithelial tumors of the salivary glands (sialomas) are uncommon. They constitute less than 5% of all salivary gland tumors in adults (1). Monographs and books in surgical pathology either do not mention these lesions or allude to them only briefly (2-4). With the exception of neurilemoma, these tumors have been reported only in the parotid gland (4). The most commonly described tumors are angiomas (hemangioma and lymphangioma), neurilemoma, and lipomas. To our knowledge, only two patients with fibrous histiocytoma arising in the parotid gland have been reported (5, 6). To these, we add a third case, of a 46-year-old man.

Case Report

A 46-year-old man sought medical attention because of a mass in the left parotid region. It had enlarged for many months before admission to the hospital. There was no pain, facial nerve involvement or disturbance of salivation. Physical examination revealed a mobile; nontender, very firm walnut-size mass. There were no other abnormalities. All laboratory data were within normal limits. A superficial lobe parotidectomy was performed during which two masses were found adherent to each other; the larger of the two occupied most of the lobe while the smaller was present in the anterior periphery of the gland. His postoperative course was uneventful.

Pathology

The specimen consisted of a portion of superficial lobe of the parotid gland measuring 4×3.5 cm in maximum dimensions. It was tan yellow and lobulated. Cut section showed a well delineated but

unencapsulated gray nodule measuring 2.8 cm in maximum dimension. Its cut surface was homogeneous and glistening. A second nodule measured 1.5 cm in maximum dimension, was firm to rubbery in consistency and was surrounded by salivary gland tissue.

The microscopic appearances of the nodules were similar. The tumor was composed of compactly packed spindle cells arranged in whorls and storiform patterns (Fig. 1). The nuclei of the cells were dense and cytoplasmic borders indistinct in many areas. Mitoses were common. No glandular, tubular, mucinous or myxoid areas were seen. Islands of parotid gland parenchyma were seen within the tumor tissue (Fig. 2). Fingerlike projections of the tumor into the surrounding tissue were present. Giant cells or foam cells were not encountered.

Follow up

About one year following parotidectomy, the patient developed papillary carcinoma of the thyroid for which total thyroidectomy and ipsilateral radical neck dissection were performed. It is now more than three years since the parotid surgery and two years since thyroidectomy. The patient is without recurrence of the parotid lesion.

Discussion

The morphologic features of the parotid tumor in this patient consisting of lack of encapsulation, interdigitation with the surrounding tissue, typical storiform (cartwheel) pattern of cellular arrangement of spindle cells, the presence of mitoses and the absence of pleomorphism, are characteristic of benign fibrous histiocytoma. These tumors are relatively common in most parts of the body particularly the skin and subcutaneous tissue. Many histologic variants are recognized: fibrous xanthoma,

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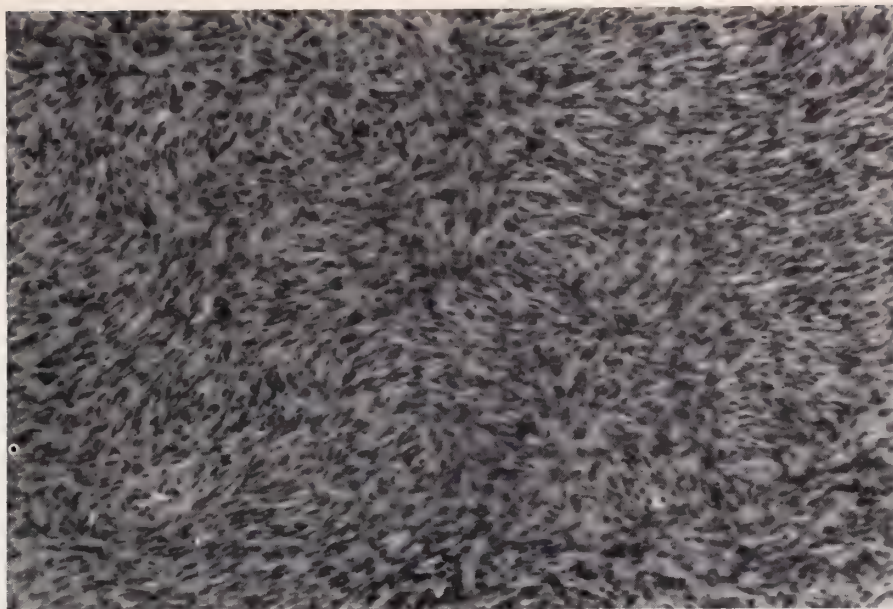


FIG. 1. Compactly packed spindle cells arranged in a storiform pattern characteristic of fibrous histiocytoma (H & E, $\times 100$).

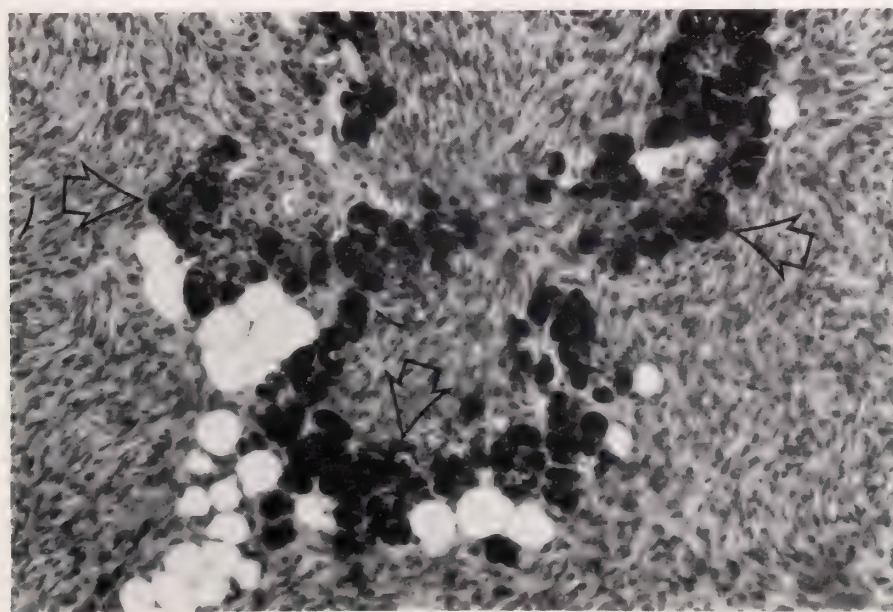


FIG. 2. Infiltration of parotid gland parenchyma by fibrous histiocytoma. Persisting acini are compressed, atrophic (arrows) and are surrounded by neoplastic tissue. (H & E, $\times 100$).

dermatofibroma, dermatofibrosarcoma protuberans, sclerosing hemangioma, giant cell tumor of soft tissue and villonodular synovitis. These various terminologies, in part, reflect the dominant histologic component of these tumors and, in part, the tissue of origin. Notwithstanding these numerous morphologic appearances, tissue cultures and ultrastructural studies have demonstrated their common derivation from the tissue histiocyte (7).

The histologic appearances of the tumor reported herein are similar to those diagnosed as a derma-

tofibrosarcoma protuberans by Junaid et al in a 50-year-old man (5). In that case, there was a recurrence of the tumor ten years after the initial resection. The morphology of both the initial and recurrent tumors were similar. Another histologic subtype, the fibrous xanthoma, was reported by Nilsen et al in a 53-year-old woman (6). There was no recurrence five months following resection. The tumor showed abundant histiocytes, in addition to which were fibroblast-like cells, xanthoma cells and scattered giant cells.

Some spindle celled lesions may be confused with the fibrous histiocytoma of the parotid gland. A pleomorphic adenoma with a predominant spindle cell component may superficially resemble the fibrous histiocytoma. In this regard, it is interesting to note that in our patient and one of the two previous reported patients (5), an initial diagnosis of mixed tumor, spindle cell type (2), was made. The accurate diagnosis of the latter may be made by reviewing multiple sections in which the epithelial components are present; the use of reticulin stains may also reveal focal epithelial pattern. The myoepithelioma is a monomorphic adenoma composed of spindle cells in a convoluted network (4). The cells are elongated and tend to be syncytial. They lack the characteristic storiform arrangement, giant cell formation and foam cells typical of fibrous histiocytomas. A distinction should also be made between nodular fasciitis and fibrous histiocytoma. The former is not a neoplasm but a lesion showing active proliferation of fibroblasts, scanty infiltration by lymphocytes and histiocytes and foci of edematous and myxoid change. The periphery of the lesion shows rich and hyperplastic vascularity. Extravasation of red blood cells is not uncommon. The lesion usually grows fast, is tender to palpation, and pursues a self limiting course.

There are too few reported cases of fibrous his-

tiocytoma of the parotid gland to permit any meaningful statements regarding its biologic behavior. Total extirpation with a wide margin can be expected to be curative (no recurrence in three years in the present case). The experience with fibrous histiocytomas of the skin and subcutaneous tissue would support this conclusion; recurrence of incompletely excised tumors is the rule.

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Intracholedochal Sodium Heparin in the Management of Retained Common Duct Stones

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Abstract

The present paper is a clinical retrospective study of the intracholedochal application of heparinized saline (20,000 units of sodium heparin in 250 ml of normal saline every eight hours) in continuous drip infusion to nine patients with retained common duct stones.

Solution of the common duct stones was verified radiologically in three patients, thus encouraging us to continue this conservative management and to propose the method in every similar case prior to surgical intervention. In addition, we want to stress the importance of continuing the treatment for at least two weeks.

One of the major problems of biliary surgery is the management of patients with retained common duct stones. The possibility of gallstones overlooked and remaining in the biliary tree after choledochotomy and common duct exploration is well established.

The incidence of retained common duct stones ranges above 2% (2) despite the continuing introduction and application of new specific techniques (6) (Fogarty catheter, fiberoptic choledochoscope) for a more effective common duct exploration.

Surgery has been, for many years, the only solution for the management of these patients. Results obtained could be considered as good but reoperations in biliary surgery are accompanied by many and difficult technical problems and a significant increase in postoperative mortality (4).

For these reasons, over the years and as early as 1891 many investigators have sought nonoperative methods for the management of these embarrassing residual calculi.

Not surprisingly voluminous literature has been accumulated in support of various conservative methods, which can be classified in two main groups:

- a) Instrumental removal of residual stones through the T tube tract (3, 12, 13).
- b) Dissolution of the retained stones by means of intracholedochal instillation of different solvents (16, 17).

The present study reports the management of nine patients with retained common duct stones by infusion of sodium heparin.

Materials and Methods

During the years 1971-1976, at the First Surgical Department, University of Athens, Greece the conservative management of nine patients with residual calculi was undertaken. These were seven females ranging in age from 35 years to 69 years with a mean age of 42.4 years, and two males 36 and 62 years of age.

All but one patient had previously undergone a removal of the gallbladder with a choledochotomy and biliary tree operative exploration. The ninth patient underwent an operative exploration of the common bile duct 14 years after a previous cholecystectomy.

The postoperative T tube cholangiography performed between the 8th and 12th day after the original operation revealed in all our patients the persistence of residual stones in the common bile duct. In one of our patients, surprising multiple common duct stones came to light.

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These patients were, initially and prior to any other procedure, treated by intracholedochal infusion of heparinized saline.

A continuous drip infusion of 20,000 IU of heparin (Sodium heparin Leo) in 250 ml normal saline was applied every eight hours. The instillation was well tolerated by the patients and there was no evidence of biliary colic or cholangitis. In addition, blood levels of bilirubin and clotting times, examined every 72 hours, remained normal. In one subject, bile leakage was seen soon after the application of intracholedochal drip which was therefore discontinued.

A new T tube cholangiography was performed on the 20th postoperative day for visualization of the extrahepatic biliary tree. The infusion was continued at least for 15 days. The duration of intracholedochal drip varied widely in our series with mean value 22 days. A repeat T tube cholangiogram was necessary for the estimation of final results.

In three of our patients, final cholangiography was interpreted as being normal, with disappearance of the previously demonstrated common duct stones and free entrance of the radiopaque media to the duodenum. With these radiologic findings, the applied conservative method was judged as successful and the T tube was removed. These

patients are now symptom-free and without any clinical, radiologic or biochemical sign of cholestasis, four months to two years after their discharge (Figs. 1-3).

Of the remaining patients, the intracholedochal infusion of heparinized saline for the management of residual stones failed since there were no radiologic findings showing the disappearance of the stones (Fig. 4).

Discussion

The fear of cholestasis and its complications, caused by retained common duct calculi stresses the absolute necessity of their management. This necessity, as well as the new trends for avoidance of reoperations in biliary surgery, because of technical difficulties, complications, and well known increased morbidity and mortality, resulted in the introduction and application of nonoperative methods for the management of these embarrassing stones.

Regarding the instrumental removal of residual common duct stones, we admit that outstanding results have occasionally been reported. In parallel, we also admit that for the application of the above methods, adequate specific armamentarium and

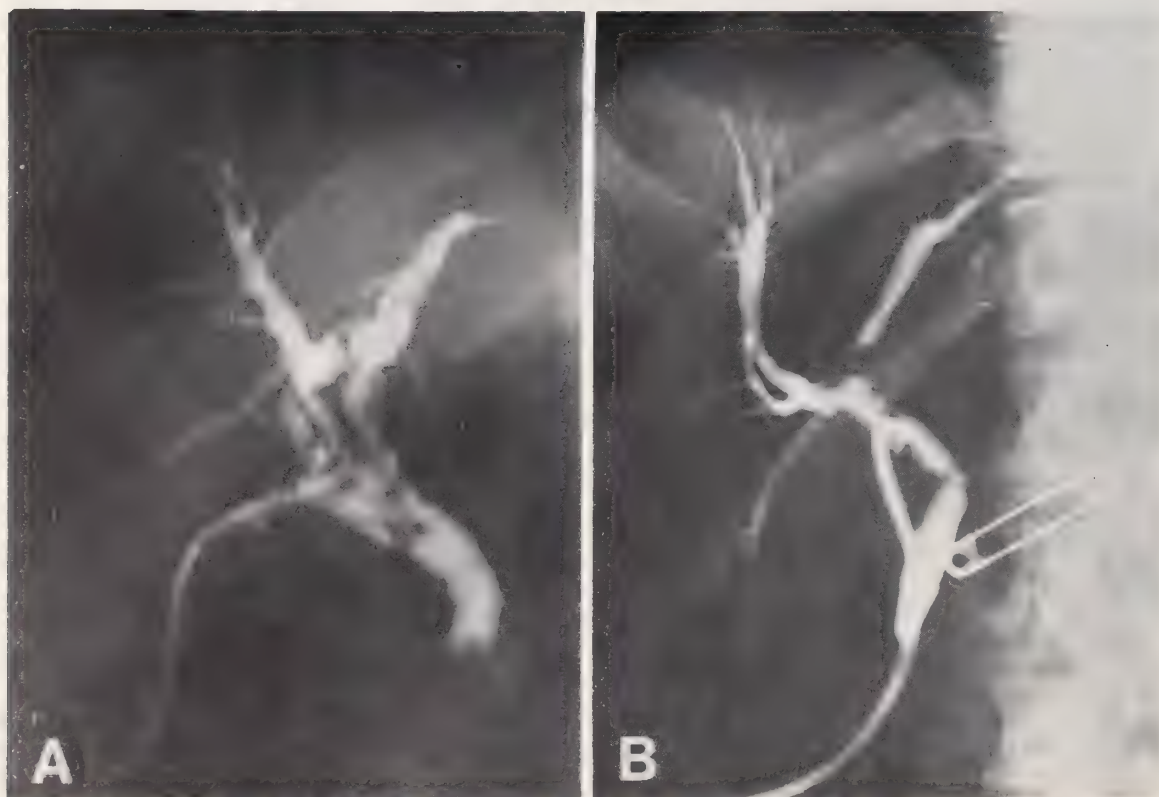


FIG. 1. (a) Obstruction of flow of dye into duodenum due to retained stone and (b) after heparin infusion.

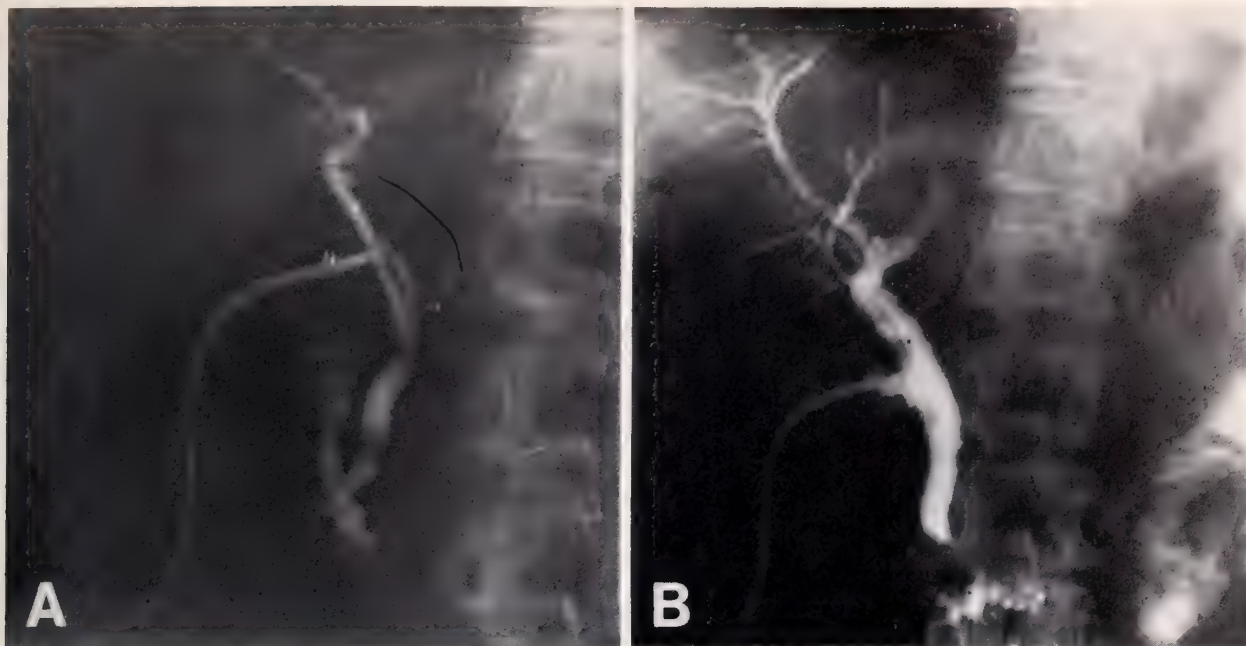


FIG. 2. T tube cholangiograms before (a) and after (b) heparin infusion.

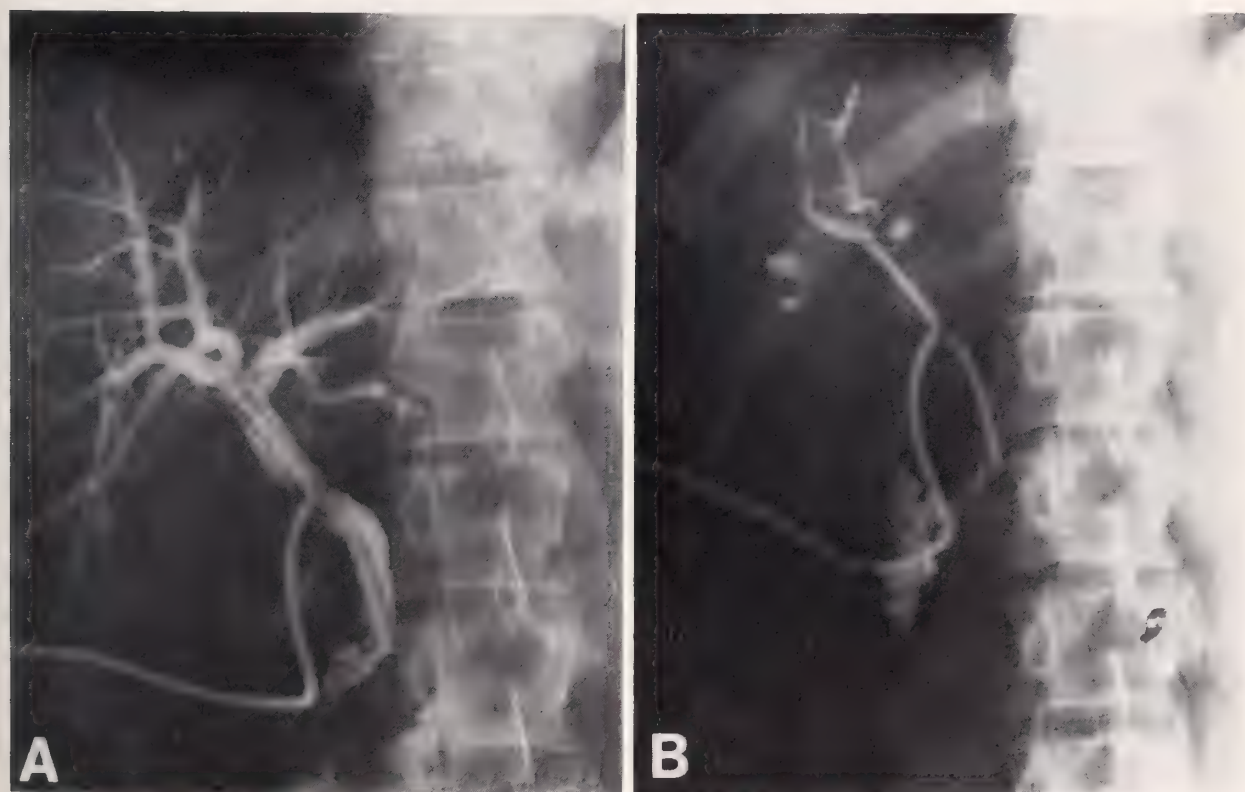


FIG. 3. This patient had two retained stones (a) that disappeared after intracholedochal heparin infusion (b).

special experience are required. In addition, a serious disadvantage of these methods are the complications seen often after its application which may be severe in rare cases (2).

The methods of intraductal infusion can be divided in two subgroups. The one deals with the mechanical irrigation of the remaining common duct calculi (5). The second one aims at the disso-



FIG. 4. Multiple retained stones unchanged after heparin infusion.

lution of the residual stones. We think that the methods belonging to the first subgroup are not very successful and are undoubtedly accompanied with complications, in spite of the occasionally good reports.

Ether (15), chloroform (1), heparin (7, 9) and sodium cholate (15, 17) are most often seen in the literature as the main solvents applied in an effort for the dissolution of the overlooked intracholedochal stones.

Sodium heparin seems to offer better results provided that side effects are simultaneously taken under consideration (10, 11). Practically this solvent, applied through the T tube is neutral as contrasted with the other substances which are accompanied with complications such as nausea, vomiting and diarrhea (2). The good tolerance of sodium heparin was also seen in our series as none of our patients experienced any complaint or any prolongation in clotting time.

It is well documented, based on previous work (10) that the effectiveness of the intracholedochal instillation of heparinized saline for the management of retained common duct stones is not due to mechanical irrigation.

Basically the theory explaining the action of sodium heparin is based on the increase in the suspension stability of bile and the increase is sustained by the addition of a highly negative charged ion (heparin) (8, 10). This action results in an increase in negative zeta potential of the cholesterol micelles

and therefore leads to their easier dispersal in the heparinized bile (14).

These biochemical descriptions of heparin's action shows why the method is applicable only for cholesterol gallstone and explains the reason for at least a percentage of failures.

Dissolution of the gallstones, which is theoretically within the capability of the method, is the ideal target. But results can be accepted as satisfactory even in cases of reduction in size of the calculi, or possibly the breaking of them into smaller particles that subsequently enter the duodenum.

This passage out of the common bile duct may be possibly helped by the continuous drip infusion acting as a flush.

Further analysis of our data, shows that the method, estimated by clinical and radiologic criteria, was successful in small percentage as contrasted with series of previous similar reports (7). In spite of this, in our opinion, the method is of great value and must always be applied and at least for 15 days prior to any further procedure, operative or non-operative, for the management of residual common duct stones.

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Thymectomy in the Adult Rat

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The thymus is an essential organ for proper development and function of the lymphoid system (1, 2). It is well known that neonatal thymectomy causes an alteration of the cellular immune response, for example, it promotes allograft acceptance and it permits tumor induction (3). The role of the thymus in the adult animal has not been clearly elucidated. It has been previously thought that its function decreases as the animal ages. Evidence, however, is coming from some research laboratories that cellular immunity is continuously under the influence of the thymus since the presence of a transplanted thymus is necessary to recover immunologic competence of animals thymectomized in the adult stage (4). Immunologic effects of adult thymectomy have been studied in various species of animals with conflicting results. Some investigators have reported little or no effect on the immune response of adult thymectomized animals, while others have reported allograft acceptance and unresponsiveness to antigens (5-12).

For these reasons, thymectomy is a useful technique in the study of the function of this organ in the adult immune system. Several methods for thymectomy of small animals, commonly employing tracheostomy and/or aspiration with suction tube, have appeared in the literature (13-17). However, detailed descriptions of these procedures are few, even for the adult rat, one of the most common animals used in research investigations. The main difficulty with this species is the close association of the thymus capsule with the pleural lining.

The following report describes the surgical procedure followed in our laboratory. Adult rats weighing 200-250 gms were used throughout the experiment.

Operative Procedure

The animal is initially anesthetized by placing it in a covered container with ether-soaked gauze pads. The anesthetized rat is secured to the dissecting board with its head toward the surgeon. Anesthesia inhalation is maintained by using a small glass beaker containing ether-soaked gauze pads. The neck and chest are depilated with an electric clipper and cleaned with an alcohol swab. A longitudinal midline incision is made on the skin from the third rib to two centimeters above the manubrium. The superficial fat is bluntly dissected to expose the pretracheal muscles, which are subsequently incised, exposing the trachea (Fig. 1). The posterior wall of the sternum is separated from the mediastinal structure using cotton-tipped applicators to prevent accidental tearing of the great vessels at the time of sternum splitting. With a small straight scissors, the sternum is split longitudinally down to the third pair of ribs by introducing one scissor blade through the entrance to the thorax. Separation of the sternum with a small retractor exposes the thymus which appears as a white bilobed organ. By means of a pair of fine forceps and cotton-tipped applicators, the thymus lobes are teased free from the surrounding tissue, starting from the poles down to their main vessels, that are usually located in the posterior side of the midsection of each lobe. These vessels are ligated with 7-0 silk. At this stage, the dissection of the lobes is temporarily discontinued. The pretracheal muscle incision is then surrounded by a loose, continuous suture of 7-0 silk (Fig. 2). The purpose of this is to prevent pneumothorax by allowing the surgeon to perform a quick closure of the operative defect. The loose suture is deflected out of the operating field and the thymus is teased completely free from the surrounding structures and removed (Fig. 3). It is important to identify and retain the lymph nodes lying just distal to the thymus.

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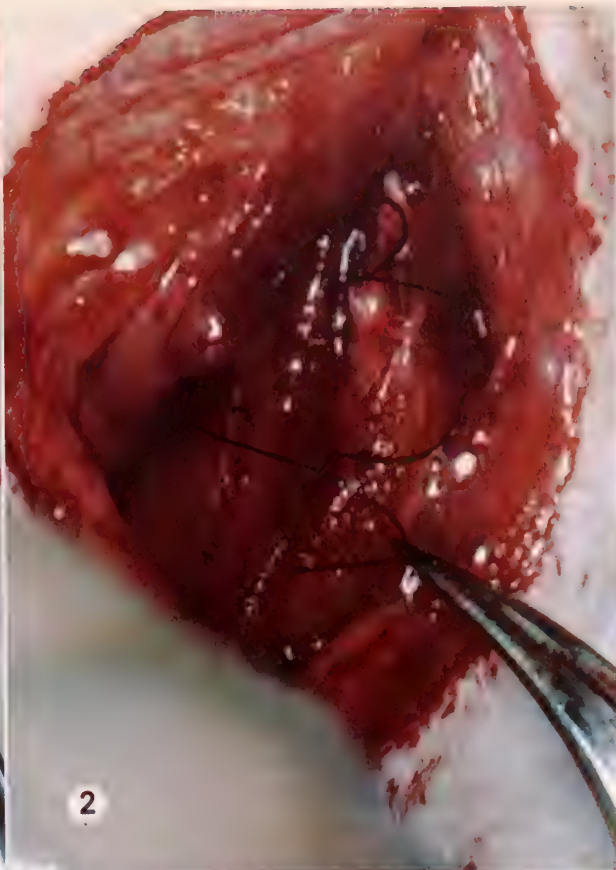
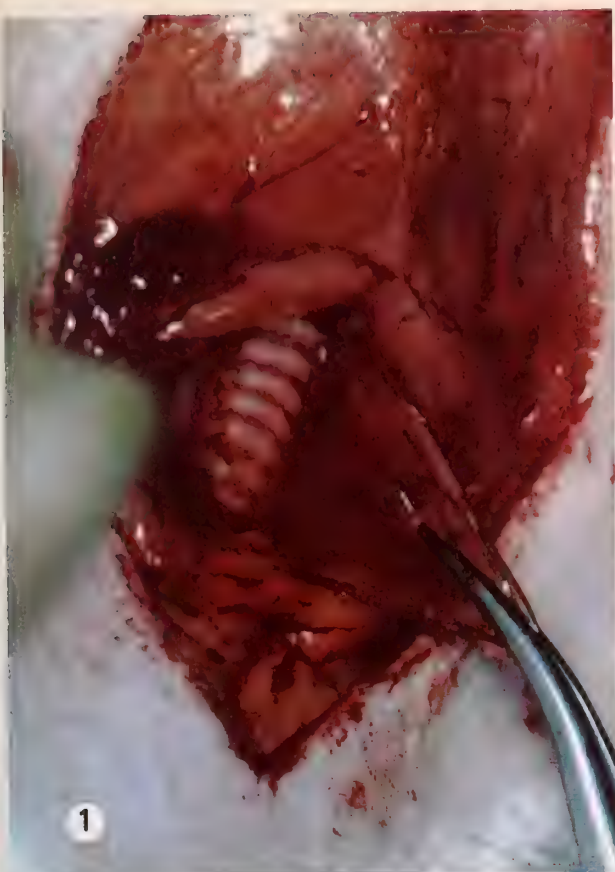


FIG. 1. After incision of pretracheal muscle and partial splitting of sternum, the trachea is exposed.
 FIG. 2. A loose suture is placed in the pretracheal muscle for rapid closure at the time of thymus ablation.
 FIG. 3. Whole thymus specimen. FIG. 4. Stitch of 4-0 silk closing the split sternum.

If these nodes are mistaken for thymic tissue and inadvertently excised, the result is uncontrollable hemorrhage. Immediate closure of the thorax is done by rapidly tightening the suture with one hand while the other hand expels the air from the chest by compression. The sternum is closed with one stitch of 4-0 silk (Fig. 4) and the skin closed with the same material. The rat is allowed to recover under a heat lamp at 35°C until normal motor control is regained. With practice, a skilled surgeon can perform the whole procedure in 15 minutes. Complete absence of residual thymic tissue is confirmed by direct inspection after the death of the animal.

A survival rate of 90% has been obtained in our hands (108 of 120) when performing this procedure precisely as described above.

Summary

A procedure for thymic ablation is described in detail. Complete thymus removal is obtained by blunt dissection alone and without resort to the common procedure of tracheostomy and aspiration with suction tube. A high survival rate depends on adhering carefully to the described procedure.

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Ureteroileosigmoidostomy: A Current Appraisal

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Abstract

Ureteroileosigmoidostomy has been performed on 116 patients during the past 25 years, primarily for carcinoma of the bladder. Eighteen patients survived longer than ten years. Significant complications occurred in 24 percent of the series and a total of 11 percent required additional surgical intervention. Hydronephrosis developed in eight patients of 94 studied. Ureteral reflux could be demonstrated by diatrizoate meglumine (gastrografin) enema in seven patients. Hyperchloremic acidosis appeared in nine cases.

Despite the frequency of supravescical urinary diversion, unqualified approval of present modalities is lacking. The number of techniques surgical ingenuity has devised over the past four decades indicates that a totally acceptable method has been elusive, since well-known deficiencies in currently employed operations have failed to completely gratify either surgeon or patient.

The operations for diversion by ureterocolic anastomosis have serious common flaws. Postoperative hydronephrosis is attributed to stenotic occlusion at the sites of communication or failure of epithelialization of the ureter within the bowel lumen. The high incidence of pyelonephritis is thought to be caused by bowel reflux up the ureters, facilitated by the difference in pressure gradients operating from bowel to renal pelvis. Hyperchloremic acidosis has occurred in all methods.

The pathogenesis of hyperchloremic acidosis has been attributed largely to two main contributing factors, operating singly or in combination. These are absorption of acid radicals through the colonic mucosa and renal injury. Kidney damage presumably results from infected hydronephrosis referable to stenosis at anastomotic junctions and the aforementioned reflux of infected material, projected by

the high intracolonic pressures through an open unhampered passageway into the kidney drainage system. When it was discovered that late hyperchloremic acidosis persisted as an unyielding complication after Leadbetter's ultimate refinement in direct ureterointestinal anastomosis, the ileac conduit became the prevailing technique of choice when ureteral diversion was found necessary.

Two distinct physiologic advantages have been claimed for the conduit. First, the urine is isolated from the fecal stream, and second, there is alleged prompt peristaltic emptying of the intestinal segment because of the absence of a sphincter. It was believed that urinary reflux was minimized, and that absorption of urinary elements was a negligible consideration since the ileal loop had no reservoir function.

Long-term results of ileal loop surgery have disclosed distinctive defects in the technique that have engendered increasing discontent. The main disadvantage of the procedure is the continual drainage of urine through the abdominal wall. Even with the best devices, ileostomy adjustment is often difficult and social rehabilitation may not be completely achieved. Stomal problems, such as stricture and retraction and paraileostomy hernia, have been unpleasant complications. In many cases, with time the conduit has lost its peristaltic capacity and abnormal distention of the loop has occurred. Long-term survivors have suffered pyelonephritis, acidosis, and disturbances in renal function.

In a comprehensive survey of 543 cases, Jaffe,

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Bricker, and Butcher (1) discovered that one third of their patients had complications. Intestinal obstruction occurred in 11.2 percent mainly from adhesions, recurrent tumor, and internal hernia. Subsequent operative procedures related to the ileal segment were performed in 26 percent of the cases, and additional surgical procedures were necessary on the upper urinary tract in seven percent.

Growing dissatisfaction with the ileac conduit has prompted the development of colonic loops, anti-reflux measures, and bladder substitutes, as well as a renewed interest in ureterosigmoidostomy. The chemical complications of ureterosigmoidostomy, its proponents claim, can be controlled by 1) large amounts of fluid intake to reduce osmotic diffusion gradients of urinary solutes and to promote frequent urination, 2) use of a rectal tube to reduce the surface area of colonic mucosa exposed for the absorption of urinary salts, 3) supplemental alkali ingestion to increase available base, and 4) augmented potassium intake.

Ureteroileosigmoidostomy is a modification of other methods of urinary diversion designed to overcome recognized disadvantages (2). The procedure involves interposition of a segment of small intestine between ureters and sigmoid, the rationale being that the isoperistaltic ileal segment will buffer the usually existing intraluminal pressure differences between colon and ureter, thus substantially reducing the hazards of reflux and infection in the upper urinary tract (Fig. 1).

Analysis of Cases

An experience of 25 years with this procedure encompasses a series of 116 patients of whom 69 were males and 47 were females (Table I). Cystectomy was performed in 84 patients. Autosture technique was utilized in 19 operations.

In 36 percent of these cases (30) the cystectomy was a second stage procedure after preliminary urinary diversion. In eight females, concurrent hysterectomy was done.

Of the entire series, 18 (of 74 cases) survived ten years or more and 23 (of 86), longer than five years (Table II). Twenty-five patients succumbed within one year of surgery while the life span of 33 was extended beyond two years following the operative procedure. Of the 37 presently known survivors, the indications for surgery in 30 was cancer. Forty-five patients died of the original disease and 24 succumbed to unrelated cardiovascular disorder.

In this series, ureteroileosigmoidostomy was necessitated by carcinoma of the bladder in 73 cases.

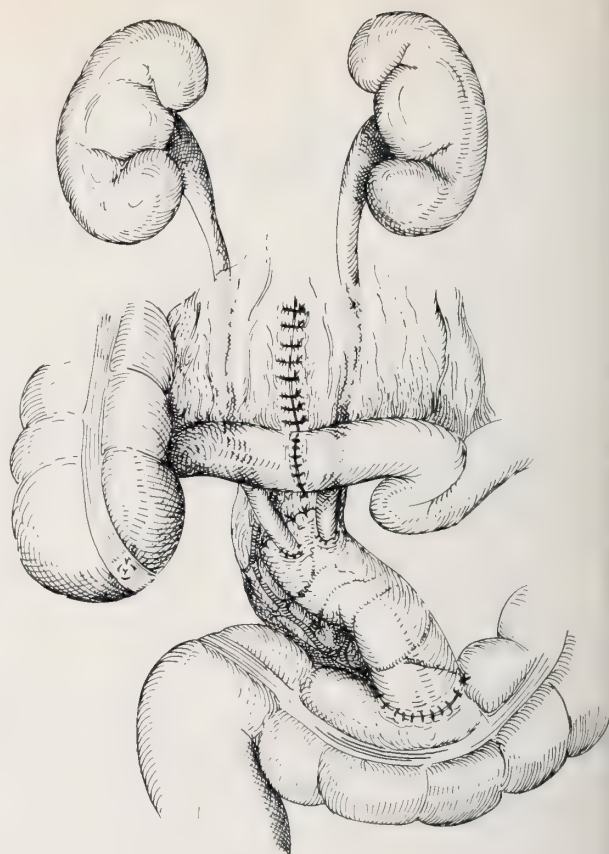


FIG. 1. Ureteroileosigmoidostomy.

TABLE I
Distribution of Cases

Number of Patients		With Cystectomy	With Hysterectomy	One Stage	Autosture
Males	69	49	—	28	12
Females	47	35	16	21	7

TABLE II
Survival

More than ten years	18 (of 74)
More than five years	23 (of 86)
More than two years	33
More than one year	20
Less than one year	25
Present known survivors	37
Died of original disease	45
Died of unrelated cardiovascular disease	24
Died after evisceration	1
Died in diabetic coma	1
Died of secondary surgical complications	2

Other indications included:

- 1) carcinoma of the prostate with invasion of the bladder (9)
- 2) rhabdomyosarcoma of the bladder (1)

- 3) inability to accept a conduit terminating in the skin-undiversion (6)
- 4) neurogenic bladder (3)
- 5) severe recurrent papillomatosis (5)
- 6) injury to solitary ureter (1)
- 7) interstitial cystitis (4)
- 8) chronic contracted cystitis (4)
- 9) carcinoma of cervix with invasion of bladder (6)

It is significant that there were six cases of undiversion because of intolerance to cutaneous urinary stoma. The youngest subject, a 20-month-old child with sarcoma botryoides, had cystectomy and diversion, and is surviving after four years, free of complication or sequella.

Complications

Significant complications occurred in 24 percent of the series (Table III). Intestinal obstruction developed in four patients. In one case, the disturbance was caused by an internal hernia in the early postoperative period; a second case of obstruction was caused by a pelvic abscess and recurrent tumor, and two other cases to adhesions. Evisceration occurred in two patients, one of whom died shortly after subsequent repair. Stenosis at the ureteroileal junction developed in three cases necessitating secondary correction. In three instances fecal fistula healed spontaneously. Since there was associated urinary leakage, it was presumed that the interruption had occurred at the ileosigmoid anastomosis. In one patient, an obstructing calculus appeared at a ureteroileal junction requiring removal. Nephro-

TABLE III
Complications (24 Percent)

Fecal fistula	Three healed spontaneously (ileosigmoid anastomosis)
Intestinal obstruction	One internal hernia One pelvic abscess and recurrent tumor Two adhesions
Calculus in kidney	Nephrolithotomy—one Nephrectomy—one (calculus pyonephrosis)
Tumor of kidney five years after procedure	Nephrectomy
Stenosis at ureterointestinal anastomosis	Three (with repair)
Evisceration in older patients	Two (repair done)
Calculus at ureteroileal junction	One (ureterolithotomy)
Hypochloremic acidosis	Nine

Eleven percent of the entire series required additional surgical intervention.

lithotomy was performed in a single case. In another patient calculus formation with attendant progressive renal damage was resolved by nephrectomy. One remarkable case concerned a 66-year-old patient who, five years after ureteroileosigmoidostomy, developed rectal bleeding that was found to be from a large papillary tumor of the renal pelvis and nephroureterectomy was carried out; there was a subsequent two-year survival.

Thus, a total of 11 percent of the entire series required additional surgical intervention.

Severe pyelonephritis occurred in ten patients, three in the early postoperative period, and the others more than six months after operative procedure. Mild pyelonephritis, amenable to biotherapy, occurred as single or repeated episodes in 16 patients. One latent diabetic had severe uncontrollable exacerbation and went into coma; he died of overwhelming electrolytic imbalance. One patient with hepatitis and two with peritonitis responded to medical therapy.

Of the 35 patients who had staged surgery, important complications occurred in only nine instances; whereas of 49 who sustained a single procedure for the combined operation, serious postoperative disturbances developed in 20. This experience corresponds to the studies of Grimes and his associates (3).

X-ray Studies

Two categories of roentgenographic follow-up were utilized (Table IV). Excretory urography was uniformly performed directly after surgery and generally at intervals of six months to a year postoperatively. To determine the effectiveness of the ileal buffer and to evaluate the frequency of ureteral

TABLE IV
X-ray Follow-up Studies

Intravenous Pyelogram-94	
Mild to moderate pelvic and calyceal distention	10
Severe hydronephrosis	8
Calculus	
kidney	5 (2 operated)
ureter	1
Chronic pyelonephritis	7 (2 pre-existing renal infection)
Normal	63 (68%)
Diatrizoate Meglumine Enemas-61	
Ureteral reflux	7
Contrast in conduit	23
Gaseous distention only (no dye)	5
Normal findings	26

reflux, enemas consisting of diluted solutions of diatrizoate meglumine (gastrografin) were performed. It was established that the force of the enema, determined by the height of the diatrizoate meglumine reservoir, had little effect on the distention of the intestinal conduit and, when present, on the intensity of subsequent ureteral reflux.

Urographic studies in 94 cases revealed relative normalcy in 63 or 69 percent of the examinations. Significant hydronephrosis was demonstrated in eight patients, and changes indicative of severe or chronic pyelonephritis were visible in seven, of whom two had a clinical history suggestive of pre-existing renal infection.

Other abnormal urographic findings included urinary calculi (5), moderate pyelectasis with mild calyceal dilatation (10), and renal tumor (1).

Diatrizoate meglumine (gastrografin) enemas were carried out on 61 patients. Ureteral reflux of varying degree was detected in seven. Contrast medium in the conduit only was present in 23 cases whereas gaseous distention of the ileal segment was clearly visible in five although no dye could be discerned. Normal results were recorded in 26 studies.

Blood Chemistry Studies

Blood chemistry examinations performed sequentially over a sufficient postoperative period to be of clinical value, were obtained from 63 patients who survived more than one year (Table V). In 40 subjects, the blood urea nitrogen level persisted at a normal standard; in 13 instances, the level fluctuated between 20 to 30 mgm percent and in ten cases between 50 to 108 mgm percent.

Normal potassium readings were constant in 47 patients. In seven cases, elevations were detected (exceeding six mgm per liter) and diminished levels (between 1.6 and 3.0 meq per liter) were found in nine.

Only six patients had hyponatremia of any significance. In eleven cases, there was marked diminution in the carbon dioxide level with a corresponding chloride elevation in nine. Lesser increases in chloride appeared in four instances.

Thus, of the 63 patients who survived more than one year on whom adequate blood chemistry stud-

TABLE V
Postoperative Blood Chemistry Studies 63 Patients (After One-Year Survival)

Results	Bun	Potassium	Carbon Dioxide	Sodium	Chloride
Normal	40	47	48	57	47
slight Elevated	13 (20-30 mgm.%)	7 (6 meq/liter)	4 (32-35 meq/liter)		4 (98-104 meq/liter)
marked	10 (50-108 mgm.%)	—	—		9 (108-115 meq/liter)
Diminished	—	9 (1.6-3.0 meq/liter)	11 (13-18 meq/liter)	6	3 (90-96 meq/liter)

ies could be obtained, azotemia and hyperchloremic acidosis were disclosed in nine. Of these, three were associated with severe pyelonephritis. Six patients required more than one hospitalization for corrective measures. In no case was death attributable to chemical imbalance, although one patient is presently on a dialysis regime for end-stage renal disease.

Conclusions

The perfect method for ureteral diversion is yet to be discovered. Assessment of all modalities presently utilized indicates that long-term dangers refer primarily to pyelonephritis secondary to reflux. It is now generally agreed that acidosis follows renal disturbances far more frequently than chemical imbalance because of acid ions traversing bowel mucosa; thus the intense interest in antireflux and antiperistaltic anastomoses to prevent progressive renal deterioration. It is our belief that the ileal conduit, used as a buffer in ureteroileosigmoidostomy, does indeed effect an acceptable measure of control against reflux and subsequent irreversible kidney damage.

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Submitted for publication December 1979.

Radiologic Notes

Claude Bloch, M.D. and Harvey Peck, M.D., Coeditors

For the reader who enjoys a challenge, the Editorial staff of the Journal is pleased to bring you an **Unknown Case**. Read the history, examine the

figures and make your own diagnosis. Then turn to p. 308 for the answer.

Submitted by Stewart Moses, M.D.

History

Two twenty-year-old patients with a long standing history of recurrent infections. You are pre-

sented with a frontal and lateral examination of the chest. One from each patient. Both have the same disease and the same clinical findings.

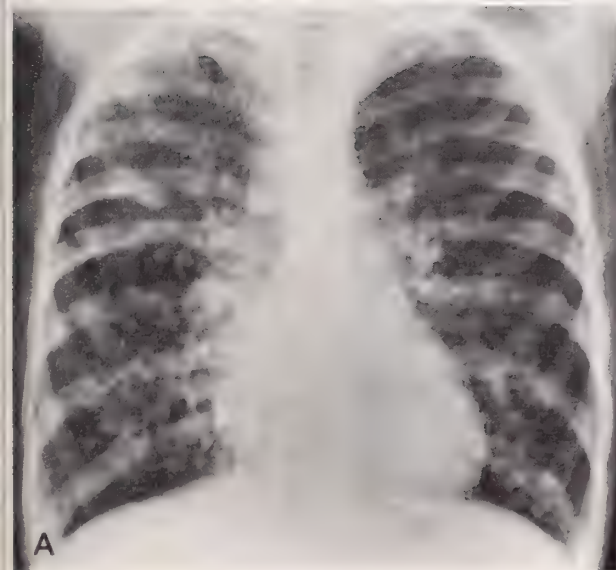


FIG. 1A

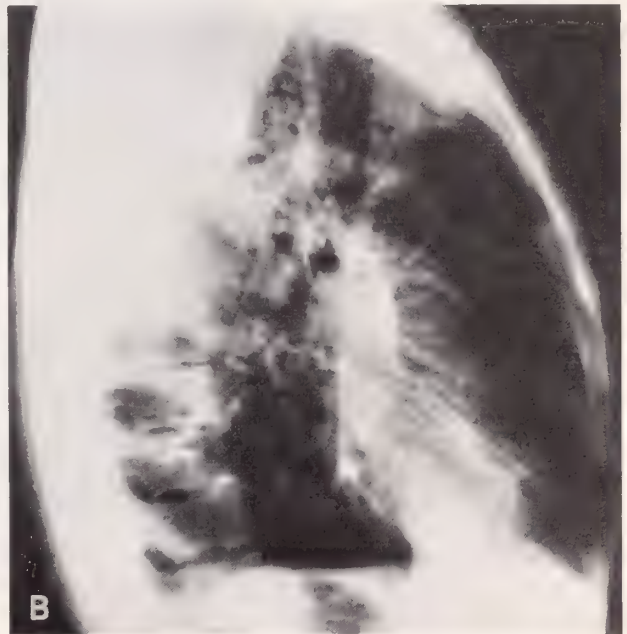


FIG. 1B

Tumor and Calculus Within a Bladder Diverticulum Case Report

ARGIL WHEELOCK, M.D. AND HANS E. SCHAPIRA, M.D.

A 66-year-old male was admitted to the hospital with a history of gross total painless hematuria. Past history disclosed multiple operations for recurrent bladder carcinoma consisting of segmental resections and transurethral procedures over an 18-year period. He also received supervoltage radiotherapy, directed to the bladder region, with a tumor dose of 4500 rads in four weeks. The patient subsequently developed a small bladder with multiple bladder diverticula. For the past few years, he had noticed the passage of many calculi.

A plain radiograph of the pelvis showed a triangular mottled calcification on the right side at the level of the ischial spine, probably situated within the right side of the bladder (Fig. 1A).

An x-ray of the opacified bladder during the course of an IVP revealed two diverticula on each side of the bladder. The stone was present in the superior right bladder diverticulum, which also

demonstrated a filling defect extending into the lumen of the bladder itself (Fig. 1B).

Under anesthesia, cystoscopy confirmed the presence of the stone and the tumor within the diverticulum. The stone was extracted and the tumor was resected. The pathology report was "transitional cell carcinoma with stromal invasion." The stone was a mixed calcium stone. The patient refused partial cystectomy and subsequent bladder biopsy was negative for tumor.

Discussion

The increased incidence of bladder cancer within bladder diverticula is well known and amounts to about 4.8% of bladder diverticula. When a bladder diverticulum and a bladder tumor coexist, the tumor most often will be found within the diverticulum. The increased incidence of bladder calculi within a bladder diverticulum is also well known, but the association of tumor and stone in the same diverticulum has rarely been reported.

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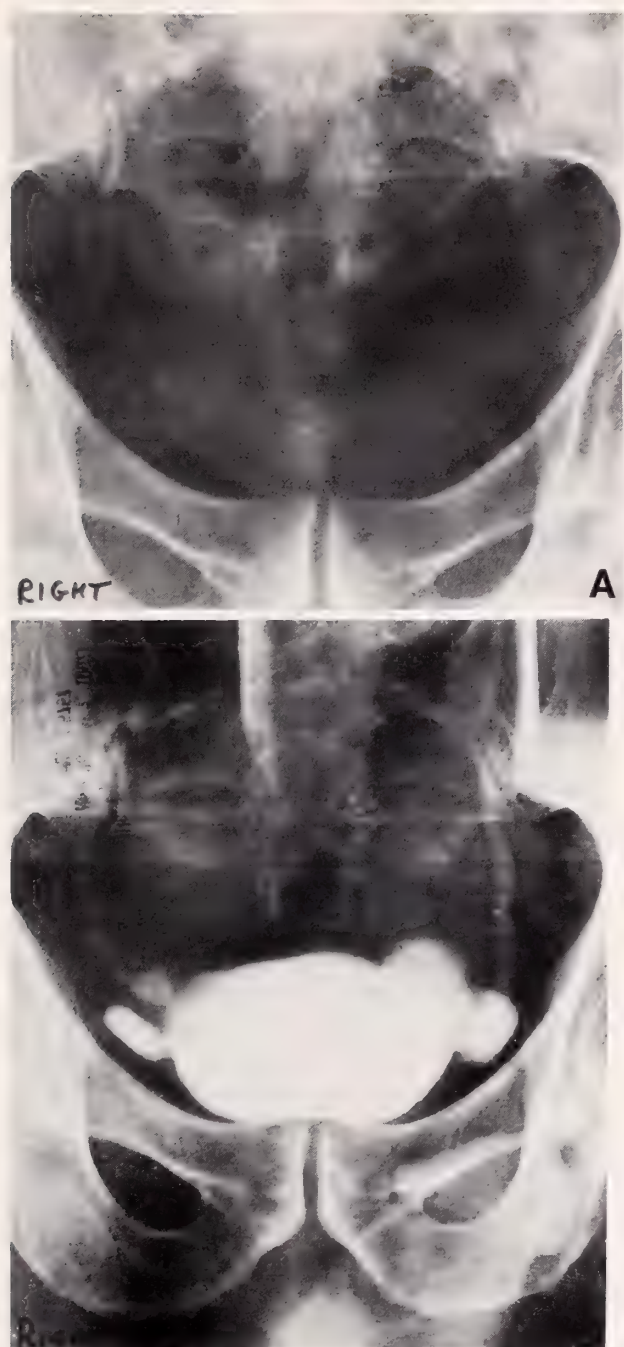


FIG. 1A. KUB showing calcification in the right bladder area.
 FIG. 1B. Cystogram showing calculus and lucent filling defect
 in right superior diverticulum.

Cont. From p. 305

Unknown Case

Answer

The major radiologic findings in these two patients are:

- a) Obstructive lung disease as evidenced by:
 - (a) flattening of both diaphragms
 - (b) an increase in the overall anterior-posterior diameter of the chest.
- b) Diffuse interstitial changes associated with medium sized cysts. Peribronchial cuffing is also evident.

The radiographic findings are consistent with what has been termed "dirty lung." The basic pathologic and radiologic findings comprising this description consist of a combination of alveolar, interstitial, cystic and bronchiectatic changes. All are associated with chronic lung disease and the major lesions that should be considered in this category are: (1) recurrent pneumonias, aspiration pneumonia, cystic fibrosis, tracheo-esophageal fistula, immunodeficiency disease, alphaantitrypsin deficiency, histiocytosis, granulomatous disease and bronchiectasis.

The findings on the chest radiograph in most aspiration disorders are quite characteristic; the pneumonic infiltrations predominantly involve both lower lung fields. Aspiration pneumonias should always be excluded even when the preliminary chest examinations fail to reveal these suggestive features. As a result, a barium esophagram should always be performed during the evaluation of any chronic lung process. For example, an H-type tracheo-esophageal fistula is a relatively uncommon congenital cause of recurrent lung disease and is only diagnosed by demonstrating the actual fistula. The latter is located high within the esophagus and extends superiorly from the esophagus to the trachea. Most other aspiration abnormalities are the result of an overflow phenomenon and are associated with either dysfunction of the swallowing mechanism or with obstructive disease of the esophagus, for example cardiospasm, esophageal carcinoma, etc.

Immunodeficiency states, either of qualitative or quantitative nature, are also associated with multiple recurrent infections within the lung and elsewhere in the body. Consequently, evaluating the electrophoretic pattern to search for abnormal globulins is essential. This was normal in the two patients presented. Alpha 1 antitrypsin deficiency is

another well recognized cause of pulmonary disease that has been associated with liver disease in both the adult and child. The pulmonary abnormalities consist of recurrent infections and investigation of this disease should not only determine the presence of serum trypsin inhibitory capacity but also the genetic sub type. Histiocytosis (eosinophilic granuloma) has a relatively characteristic pattern. The upper lung fields are predominantly involved and cystic changes that impart a diffuse honeycomb appearance to the lung are noted in the more advance cases. Recurrent infections however are usually not the main feature of the disease despite the pulmonary appearance.

In any young patient or young adult who presents with recurrent episodes of tachypnea associated with evidence of air trapping, atelectasis and pneumonias, it is imperative to consider cystic fibrosis as the underlying cause. Cystic fibrosis is inherited by autosomal recessive transmission and is more common in caucasians than blacks. It is a disease that manifests a wide degree of severity and is associated with a generalized dysfunction of the exocrine glands. As a result, abnormal mucous secretions are produced which obstruct the ducts of glandular and organ systems throughout the body. Chronic pulmonary disease is a well known complication. The predominant bacterial flora cultured from the lungs and nasopharynx are staphylococcus aureus and pseudomonas aeruginosa.

Extra pulmonary abnormalities are usually present and pansinusitis, malabsorption and intestinal obstruction (meconium plug equivalent) form the major findings.

In the newborn period, meconium ileus or rectal prolapse are presumptive evidence for the disease. The diagnosis is confirmed by an abnormally high sodium in the sweat.

The onset of pulmonary disease is variable and may appear days, weeks, months or years after birth. Bronchial and bronchiolar obstruction by abnormally tenacious secretions is the primary manifestation and is followed by widespread bronchial infection. The pulmonary disease if not checked by antibiotic and other therapy, is frequently severe and progressive. The usual radiographic progression is for repeated pneumonias superimposed upon a picture of segmental and lobar atelectasis in a hyperinflated lung with streaky infiltrations. Eventually chronic changes supervene and a reticular nodular pattern results with areas of "honeycombing" and patchy atelectasis and/or consolidation. In addition extensive air trapping develops secondary to mucous plugging. Large areas of

cystic change and multiple peribronchiolar abscesses are seen frequently. Eventually, with progression, severe emphysema and cor pulmonale develop.

Patients over the age of 15–20 manifest a better prognosis than younger patients. Their survival, in all probabilities, depends upon a relatively mild degree of pulmonary disease. Males also demonstrate a better prognosis than females; older patients, however, manifest a greater incidence of

certain complications—sinusitis, nasal polyps, abdominal obstructive symptoms and spontaneous pneumothorax.

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Seminars in Reproductive Medicine

Norbert Gleicher, M.D., Editor

Phakomatoses in Reproductive Medicine*

NORBERT GLEICHER, M.D., CHARLES T. MILANO, M.D., ALLAN E. RUBENSTEIN, M.D., AND LIANA DELIGDISCH, M.D.

Introduction

The phakomatoses represent a group of poorly understood and probably underrecognized genetic disorders which, in their classic forms, have a number of common appearances, involving mainly gross deformities of skin, bone, and nervous tissue (1). The most commonly encountered phakomatoses are listed in Table I. The name is derived from the Greek PHAKOS, a word with multiple meanings, one of which is "birthmark." This group of disorders has been attended to in the past primarily by neurologists and dermatologists. In spite of their genetic etiology, and in spite of the recent recognition that phakomatoses represent multisystem processes, little or no attention has been given to reproductive aspects of this group of disorders. Investigating the literature, we were, in fact, unable to locate a single publication covering the subject specifically.

Our interest in the reproductive aspects of phakomatoses was triggered by the unusual gynecologic presentation of such a case that was reported in more detail elsewhere (2). A short summary will be presented here, only to demonstrate the possible

reproductive repercussions, which phakomatoses may exhibit on the life of afflicted women. It is our belief that with increasing awareness more of these cases will be diagnosed. A larger variety of reproductive pathology should then be recognized. At present, only very limited information is available. It is the purpose of this seminar to review gynecologic and obstetrical aspects of phakomatoses, with the special intent of demonstrating, that the reproductive organs may be affected similarly to all other organ systems.

Case Report

A 31-year-old nulliparous Jamaican female was admitted to the gynecologic department of The Mount Sinai Services at Elmhurst General Hospital because of uncontrollable vaginal bleeding.

She was diagnosed as having Marfan's Syndrome twenty years prior to her admission. Progressively enlarging skin nodules were first noted during her adolescence. At age 15, she underwent surgical reduction of her elongated left lower extremity. Her elongated left upper extremity and a very severe kyphoscoliosis were left uncorrected. Other deformities, which increased in severity from adolescence on, included large varices of her distal extremities, digital contractures and laxities, multiple fleshy nontender skin nodules, scattered hypopigmented macules, patches of hyperpigmentation, and marked cervical (uterine) hypertrophy. An extensive work-up revealed these additional abnormalities: the hematocrit was 24.2 percent, the hemoglo-

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Presented at the weekly staff conference of the Department of Obstetrics and Gynecology, The Mount Sinai Medical Center, New York, on November 21, 1979.

bin was 6 gram percent. X-ray of the spine showed severe kyphoscoliosis of the upper lumbar and lower thoracic and cervical spine and levoscoliosis of the lumbar spine. The long bone roentgenographs showed a sclerotic island in the upper third of the left humeral shaft and both fibulae. Serpentine-like defects of the distal third of the esophagus and within the stomach in the upper GI series, were interpreted as probable varicosities.

A total abdominal hysterectomy and left salpingo-oophorectomy were performed. Unique uterine pathology was noted (2). The endometrium

showed diffuse decidual transformation of the stroma and atrophic endometrial glands; a picture consistent with prolonged oral contraceptive therapy which this patient received for severe uterine meno-metrorrhagia.

The most striking feature was the thickening and marked tortuosity of blood vessels, mainly the spiracular arterioles. The walls of these blood vessels exhibited a marked increase in both smooth muscle and elastic fibres, and a large number of vasa vasorum could be seen in the adventitia. Granular lumina showed numerous calcifications (Fig. 1).

The vascular pathology within the uterus was similar to the vascular pathology noted in other organs. Similarly, calcifications were noted on soft tissue x-rays in a number of other organs. For more detail and photographic description of this patient, the reader is referred to another publication (2).

At this stage it had become apparent that this patient did not suffer from Marfan's syndrome, as she had been made to believe in the past, but the correct diagnosis for this unique clinical picture was still unknown. It was the accidental visit to the very popular Broadway Play, "The Elephant Man" by one of the authors, that finally led to the diagnosis.

TABLE I
Phakomatoses

	Inheritance	Incidence per births
Von Recklinghausen disease (neurofibromatosis) (NF)	autosomal dominant	1/2,000
Tuberous sclerosis (TS)	autosomal dominant	1/150,000
Klippel-Trenaunay-Weber (KTW)	sporadic disorder	extremely rare
Von Hippel-Lindau	autosomal dominant	1/13,000
Sturge-Weber	represents the cerebral form of KTW	

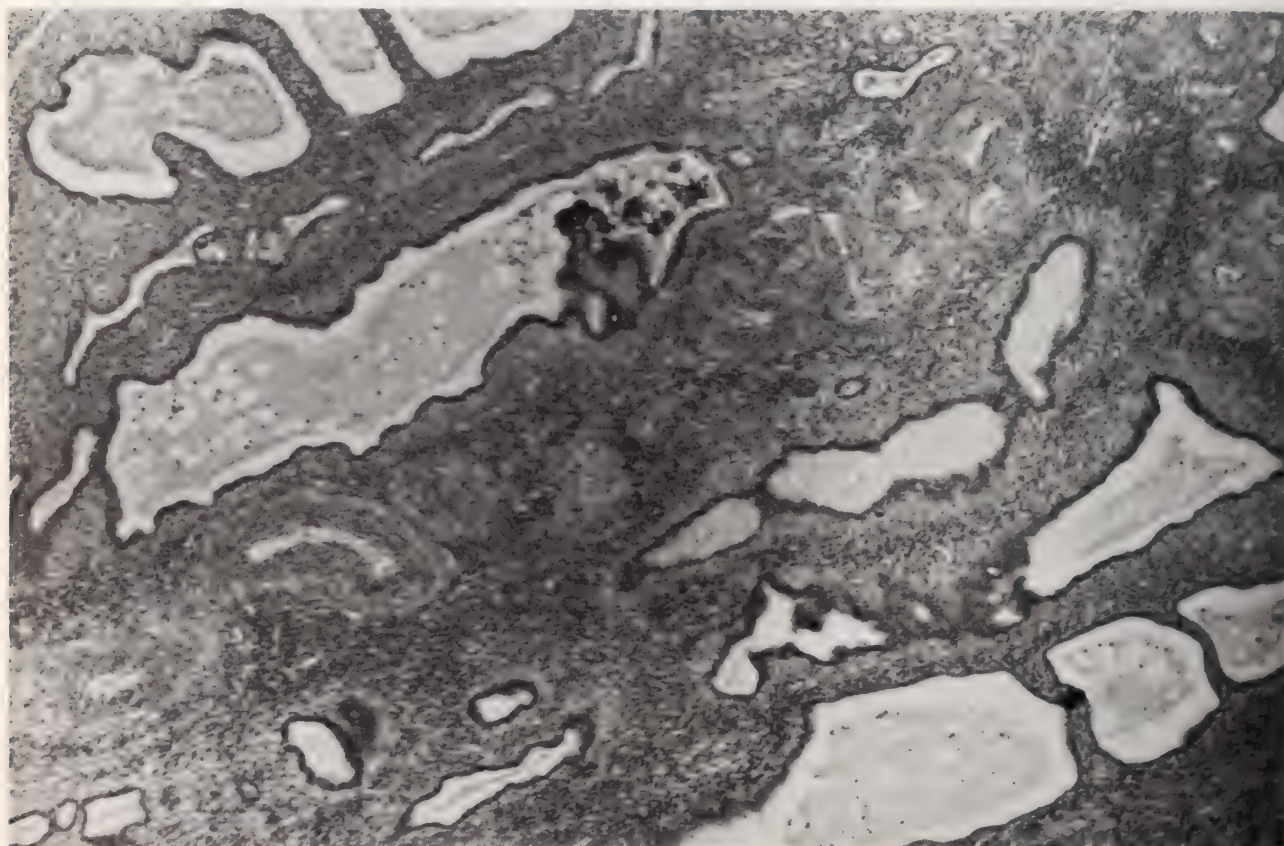


FIG. 1. *Uterine histology*: Note the tortuous blood vessels with markedly thickened wall seen next to an endometrial gland with atrophic epithelium and intraluminal calcifications. Hematoxylin-Eosin $\times 40$.

The similarity of the patient's and the Elephant Man's appearance, as projected during the play (Fig. 2), was convincing enough to be followed up in the library. It was Frederick Treves' original report from 1885, published in *The Transactions of The Pathological Society of London* under the title "A case of congenital deformity" (3) which led us to consider a phakomatosis as the correct diagnosis for our patient. In reviewing the literature it became very quickly apparent that Dr. Frederick Treves had been correct in diagnosing John Merrick as suffering from neurofibromatosis. Our patient, on the other hand, was obviously afflicted by one of the phakomatoses but none of the textbook descriptions was completely satisfactory. The most similar case was described by Graul in the German literature and was termed Klippel-Trenaunay syndrome, now called Klippel-Trenaunay-Weber (KTW) (4). Because additional symptoms more typical for other phakomatoses were also present in our patient we reported the patient as a case of "mixed phakomatosis" (2).

Once this diagnosis was made, it finally became possible to obtain adequate expert consultation. The patient was consequently reevaluated by a neurologist, which led to the conclusion that this patient suffered from an extraordinary combination

of two rare congenital disorders, tuberous sclerosis (TS) and Klippel-Trenaunay-Weber (KTW). The unique neurologic aspects of this case will be discussed elsewhere.

Discussion

The case presented here, clearly demonstrates the prevailing ignorance, even in medical circles, concerning an uncommon group of disorders, the phakomatoses. Had it not been for Dr. Treves' scientific documentation of John Merrick's plight (3) and its most astonishing recent revival on stage (5) our patient might still be falsely considered a case of Marfan's syndrome.

More important is the recognition that phakomatoses may give rise to pathology in all organ systems, including those of reproduction.

Gynecological Aspects

Because of their striking appearance in their classical forms, gynecologic presentations of phakomatoses have until recently been almost wholly neglected. Initially thought to involve only tissues of ectodermal origin, thus not affecting the reproductive system, most of the genital tract pathology in phakomatoses has been reported as incidental autopsy findings. However, due to the work of a number of investigators (1) the phakomatoses are now recognized as multisystemic genetic disorders involving diffuse malformations of several germ layers.

NEUROFIBROMATOSIS (NF)

Von Recklinghausen neurofibromatosis (NF) is an autosomal dominant disorder with an incidence of 1/2000 births. It classically is seen with 1) multiple cutaneous macular pigmented areas (cafe-au-lait spots), 2) multiple dermal neurofibromas (benign tumors of neural origin), and 3) a propensity to benign cerebral tumors, particularly acoustic neuroma, optic glioma (in children) and meningioma. Cafe-au-lait spots are usually present at birth or appear in infancy. Dermal neurofibromas usually develop around puberty. It is also recognized that visceral lesions may occur in association with NF. Frequently associated gynecologic pathology is most probably unrelated to the disease itself and represents only incidental association. Such pathology includes leiomyomata of the uterus (fibroid tumors), ovarian cysts, and carcinomas (1). The hallmark lesion of NF, the neurofibroma, has



FIG. 2. *The Elephant Man* as shown in reference 3.

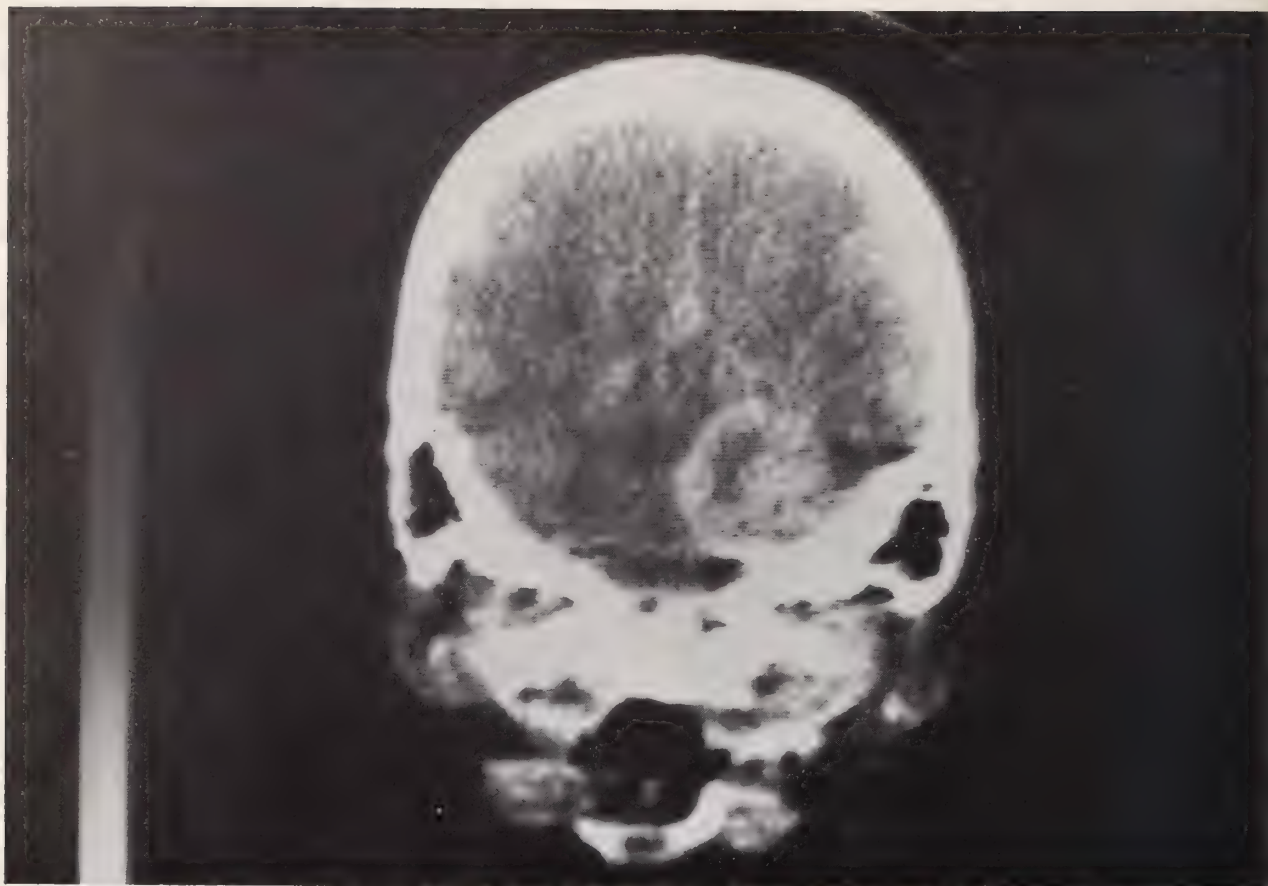


FIG. 3. CT-scan of brain: Note the large acoustic neuroma in this pregnant patient. For details see text.

been reported in uterus, ovary, broad ligament, and vagina (in descending frequency), with an overall frequency of up to 17 percent (1). Although usually asymptomatic, neurofibromas may be present as an abdominal or pelvic mass, or may cause dysmenorrhea and/or pelvic pain.

Macroscopically, neurofibromas are soft and slightly elastic in consistency, with whitish-yellow appearance. They may undergo cystic degeneration, particularly if ovarian in location. Malignant features, such as extension into mesosalpinx or into the presacral lymph nodes has been reported. It is estimated that up to ten percent of genital neurofibromas exhibit frank sarcomatous changes (1). This percentage is possibly similar to the incidence of sarcomatous degeneration of neurofibromas in other than genital sites.

Microscopically, the predominant picture is hyalinization of blood vessel walls with peripheral collagen sclerosis and varying quantities of precollagenous fibres.

Adjacent organ involvement may also be of importance to the gynecologist or pelvic surgeon. Neurofibromas and ganglioneuromas may develop in

the gastrointestinal tract. The caecum and large bowel are the most common sites of involvement. Neurofibromas of the appendix, mesentery, and in the retroperitoneum may appear as lower abdominal or pelvic masses. Neurofibromas have also been reported in ten percent of autopsy proven cases in the urinary tract, involving urethra, bladder, ureter and kidneys (1).

Sturge-Weber Syndrome

This represents the cerebral form of the KTW syndrome, which previously had not been association with specific gynecologic pathology. Sturge-Weber is characterized by capillary or cavernous hemangiomata within the cutaneous distribution of the trigeminal nerve and by venous hemangiomata of the leptomeninges.

The CNS lesions lead to seizures, intracerebral calcifications and atrophy of the cerebral cortex. Its visceral lesions have been poorly described but are reported to involve the genital tract in up to 15 percent of all cases. The most common gynecologic presentations are menometrorrhagia, secondary to hemangiomas of the uterus, followed by acute hem-

operitoneum secondary to ruptured ovarian hemangiomas. When the pituitary gland is involved, a so-called "adiposogenital syndrome" has been reported, which occurs with obesity and hirsutism (1). Grossly, dark-red spongy masses are found, that on histologic examination, represent localized angiomas with hyalin degeneration, focal dilatation and calcification, and eventually lead to transformation of vessels into laminated concretions.

To our knowledge, no case of uterine or ovarian angioma transforming into an angiosarcoma has been reported to date.

Tuberous Sclerosis (TS)

TS, also known as Bourneville's disease, is an autosomal dominant phakomatosis that is clinically manifested by the triad of seizures, mental retardation and adenoma sebaceum. Gynecologically, these patients have a high incidence of hypoplasia or agenesis of the uterus and ovaries. Recently, several cases of precocious puberty, secondary to tuberous involvement of the hypothalamus have been reported (1).

Our patient exhibited some minor signs of TS. They included: 1) multiple cutaneous abnormalities, including subungual fibromas, a shagreen patch, hypopigmented macules and a cafe-au-lait spot, 2) sclerotic islands of the long bones, 3) a family history of two affected relatives, and 4) an asymptomatic calcified lesion of the brain, suspected on routine skull x-ray, and confirmed by computerized axial tomography.

Such lesions consist of proliferating glial elements and ganglion cells and are often calcified. "Tuberous" is derived from the "potato-like" appearance of the cerebral lesions.

Von Hippel-Lindau Disease

This syndrome represents an autosomal dominant phakomatosis with incomplete penetrance and variable expression, estimated to occur in up to 1/13,000 newborns. The hallmarks of this disorder are hemangiomatous lesions of the cerebellum and retina, and cysts of the kidneys and pancreas. Similar lesions have also been described in ovaries, uterus and vagina, but are rare (1).

Other Phakomatoses

The list of phakomatoses in which multisystem and specifically gynecologic pathology can be found, has steadily increased. Syndromes with reported gynecologic pathology include the Gruber syndrome and the De Sancti syndrome (1). It seems

reasonable to assume that additional visceral components, including those of reproductive organs, will be described, as the recognition of the phakomatoses, a still neglected and poorly understood group of disorders, will improve.

OBSTETRICAL ASPECTS

The number of phakomatoses afflicted patients is steadily increasing as more cases of this group of disorders are clinically recognized. Genetic counseling, prenatal diagnosis and finally embryologic research into the etiology of phakomatoses will undoubtedly increase and improve, as the scientific interest in these disorders advances.

Only the most frequently occurring phakomatosis, NF, has been investigated to some extent in association with pregnancy. Several studies have noted that pregnancy worsens the peripheral manifestations of NF (6). Six cases of acoustic neuroma in non-NF patients, seen in the last trimester of pregnancy, have previously been reported (7). Figure 3 demonstrates the CT-scan of a 26-year-old woman with three cafe-au-lait spots who recently came to our obstetrical service at approximately 30-weeks gestational age. An enormous acoustic neuroma can be recognized.

The reasons for the worsening of the clinical picture of NF during pregnancy are still poorly understood. Fluid accumulation in the maternal tissue compartments is considered one factor, but others may be involved, particularly in view of a recently observed maternal effect on the severity of NF. Miller and Hall, analyzing 62 NF patients, reported that a maternal history of NF caused more severe disease in afflicted children than a paternal history of NF (8). These findings led the authors to suggest that in addition to the maternal gene for NF, which the fetus inherits, a product of expression of this gene may be produced during pregnancy and be passed across the placenta, influencing disease.

Nerve growth factor (NGF) is a protein that is believed to be responsible for the growth and the differentiation of neural crest derived cells. It is considered to represent one of a number of growth factors that may be involved in the pathogenesis of NF. Fabricant and coworkers demonstrated elevated NGF levels in the central form of NF, familial bilateral acoustic neuroma (9). We were able to confirm at our institution that NGF was elevated in sera from patients with NF who had central nervous system involvement (10).

We were also able to identify NGF in normal human amniotic fluid and are presently investigating its distribution in amniotic fluid throughout

pregnancy. It is our purpose to establish normal values for the different stages of pregnancy in order to be able to investigate if fetuses with NF exhibit elevated levels. Such an approach may in the future allow the early prenatal diagnosis of NF. This work is done by collaboration between investigators from the Departments of Neurology and the Department of Obstetrics and Gynecology at The Mount Sinai Hospital.

Summary

We reviewed the gynecologic and obstetrical aspects of phakomatoses that we consider to be of relevance for the practice of reproductive medicine. It may be concluded that both aspects are in urgent need of further investigation. The phakomatoses represent a widely underrecognized group of disorders which, if adequately appreciated in their incidence, will achieve increasing importance to the practice of reproductive medicine.

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Seminars in Physiology

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Regulation of Pathways of Alcohol Metabolism by the Liver

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Abstract

The major pathway for alcohol (ethanol) metabolism by the liver is the alcohol dehydrogenase pathway. This pathway is regulated by the transfer and reoxidation of reducing equivalents from NADH into the mitochondria as well as the activity of alcohol dehydrogenase itself. A minor pathway for ethanol metabolism involves the ability of isolated microsomes to oxidize alcohols in a reaction mediated by cytochrome P-450. This microsomal pathway is induced after chronic ethanol consumption. Induction of this pathway may contribute to the adaptive increase in ethanol metabolism found after chronic ethanol consumption. Recent studies suggest that the microsomal pathway may involve the interaction of ethanol with hydroxyl radicals generated during microsomal electron transfer.

Introduction

The liver represents the main site of oxidation of ethanol. The activity of major enzyme systems associated with ethanol metabolism are found to be highest in this organ so that the extrahepatic oxidation of ethanol does not generally exceed 10% to 15% (1, 2). Ethanol is oxidized to acetate in a two-step reaction involving the enzymes alcohol dehydrogenase and aldehyde dehydrogenase. Acetate produced from the oxidation of ethanol in the liver is released into the blood and oxidized in peripheral tissue (3, 4). The end products of ethanol metabolism in the liver represent the end products of acetate metabolism and include CO₂, fatty acids and cholesterol. During the oxidation of ethanol to acetate, acetaldehyde is produced as an intermediate. Most of the acetaldehyde produced by the

oxidation of ethanol in the liver is efficiently oxidized within the mitochondria (5) as levels of acetaldehyde are three orders of magnitude lower than the concentration of ethanol (6, 7). Some acetaldehyde may be released into the blood; although liver levels, circulating levels, and extrahepatic levels of acetaldehyde are low, acetaldehyde is considerably more reactive than ethanol and there have been suggestions that acetaldehyde may play a role in the actions of ethanol (8).

Alcohol dehydrogenase and aldehyde dehydrogenase utilize NAD⁺ as a cofactor, with the subsequent production of NADH. As a consequence of ethanol metabolism, the liver NAD⁺/NADH redox ratio is markedly reduced. Actually, because of binding of pyridine nucleotides, the free NAD⁺/NADH ratio cannot be measured. Instead, the ratio of oxidized and reduced metabolites, which are substrates or products of a reaction catalyzed by an active NAD⁺-dependent dehydrogenase, is measured (9, 10), that is, the lactic dehydrogenase reaction; pyruvate + NADH $\rightleftharpoons$ lactate + NAD⁺. The NAD⁺/NADH ratio can therefore be expressed as the pyruvate/lactate ratio. Since lactic dehydrogenase is a cytosolic enzyme, the pyruvate/lactate ratio reflects the redox state of the cytosolic compart-

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ment of the liver cell. The acetoacetate/ β -hydroxybutyrate ratio reflects the redox state of the mitochondrial compartment. In the presence of ethanol, the pyruvate/lactate and acetoacetate/ β -hydroxybutyrate ratios are reduced. Many of the metabolic effects of ethanol, that is, inhibition of gluconeogenesis (11), impaired oxidation of fatty acids (12, 13) and decreased activity of the citric acid cycle (14, 15) can be attributed to the ethanol-induced reduction of the hepatic redox state (16, 17).

The Alcohol Dehydrogenase Pathway of Ethanol Metabolism

Alcohol dehydrogenase is a metalloenzyme found in the cytosol of the liver cell. Although present in other tissues such as kidney, stomach and the intestinal tract (18, 19), greatest activity is found in the liver cell and the major portion of ethanol

metabolism is attributed to it. Oxidation of ethanol by alcohol dehydrogenase generates NADH in the cytoplasm; reoxidation of NADH may be rate limiting for the overall metabolism of ethanol (20, 21). Compounds that oxidize NADH, such as pyruvate, D-glyceraldehyde, methylene blue and fructose, increase the rate of ethanol metabolism. Normally, cytoplasmic processes appear insufficient to reoxidize all the NADH generated by ethanol metabolism, consequently, mitochondrial oxidation of the reducing equivalents of NADH is required. Because intact mitochondria are impermeable to NADH (22), reducing equivalents are transported into the mitochondria via substrate shuttles. Three major shuttle mechanisms for the transport of reducing equivalents into the mitochondria have been proposed. These are 1) the α -glycerophosphate shuttle, 2) the malate-aspartate shuttle and 3) the fatty acid elongation shuttle (Fig. 1) (23-27). Essentially, the

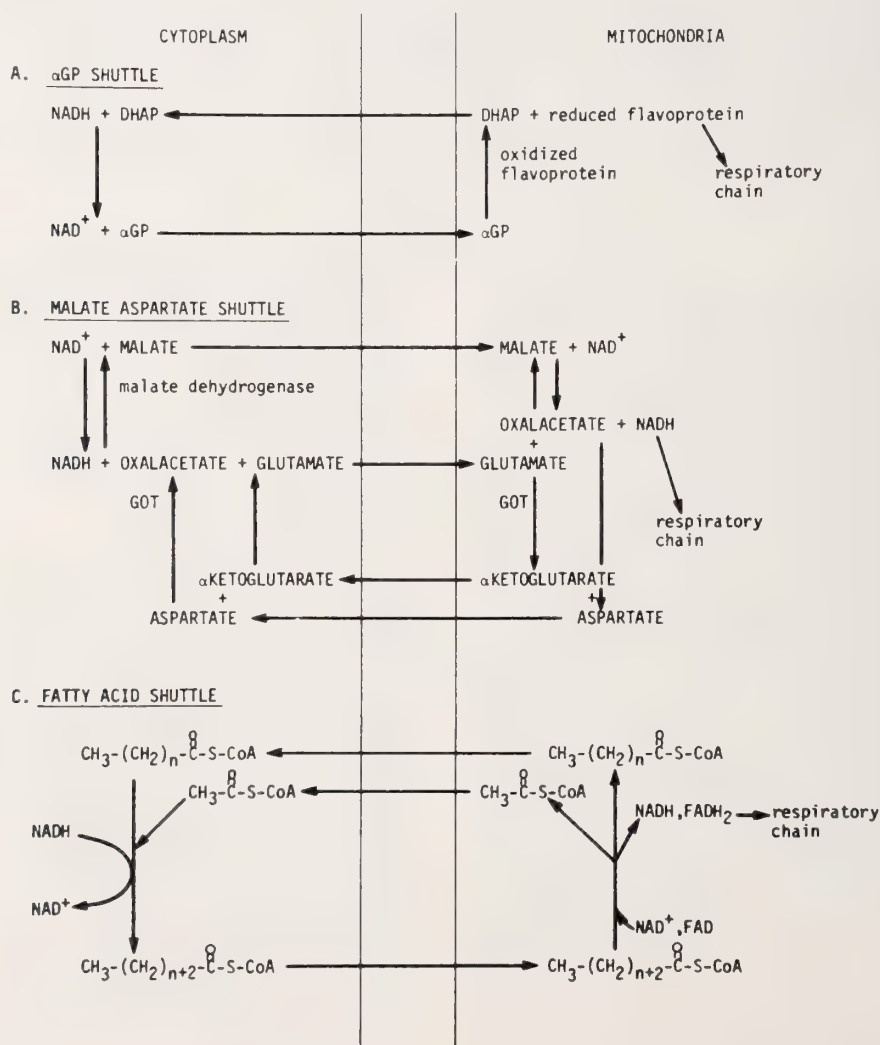


FIG. 1. Shuttle mechanisms for the transfer of reducing equivalents from NADH into the mitochondria.

shuttles involve reoxidation of NADH coupled to the reduction of either dihydroxyacetone phosphate, oxalacetate or the carbonyl acyl group during elongation of a fatty acid. The reduced metabolite is transported into the mitochondria where it is reoxidized at the expense of intramitochondrial NAD⁺ or FAD. The NADH and FADH₂ produced intramitochondrially is reoxidized by the mitochondrial respiratory chain. The oxidized metabolite leaves the mitochondria where it is available for the next round. In view of the restricted permeability of the mitochondrial membrane to oxalacetate, the malate-oxalacetate shuttle as originally proposed (27, 28) required the following modification; oxalacetate is transaminated to form aspartate, which then leaves the mitochondria (26, 27). In general, the shuttles are regulated by the activities of the cytosolic and mitochondrial enzymes which comprise the shuttle, for example, α -glycerophosphate dehydrogenase, malate dehydrogenase, aspartate aminotransferase (glutamic-oxalacetic transaminase), and by the ability of the oxidized or reduced metabolites to transverse the mitochondrial membrane. All three shuttles have been reconstituted in *in vitro* systems, using isolated liver mitochondria and the extramitochondrial components of the shuttles (29–34).

In view of the above considerations, the activity of the alcohol dehydrogenase pathway of ethanol metabolism can be affected by a) the activity of alcohol dehydrogenase; b) the activity of substrate shuttle systems transporting reducing equivalents into the mitochondria and c) the ability of the mitochondrial respiratory chain to reoxidize the reducing equivalents.

Regulation of the Alcohol Dehydrogenase Pathway in Isolated Hepatocytes

In studies with isolated systems such as liver slices (35), perfused livers (36) or hepatocytes (37–39), the rate of ethanol metabolism was shown to be limited by the transfer and reoxidation of reducing equivalents by the mitochondrial respiratory chain rather than by the activity of alcohol dehydrogenase. For example, in isolated hepatocytes from male Sprague-Dawley rats, pyruvate (acting as an NADH-oxidizing agent) increased the rate of ethanol metabolism by 153% (39), suggesting that cofactor availability, that is, the reoxidation of NADH to NAD⁺, and not the activity of alcohol dehydrogenase, was rate limiting for ethanol oxidation in these preparations. However, it is not clear whether the rate-limiting step in ethanol oxidation reflects the transfer of reducing equivalents

into the mitochondria as catalyzed by substrate shuttles, or the ability of the respiratory chain to reoxidize the reducing equivalents. The concept that the concentration of substrates which participate in the malate-aspartate shuttle play an important role in ethanol metabolism has been suggested by Williamson and collaborators (37, 38). In the fasted state, in which the content of shuttle substrates is presumably low, ethanol oxidation may be limited by the activity of the shuttles. In the fed state, in which the content of shuttle substrates is presumably sufficient, ethanol oxidation may be limited by the capacity of the mitochondrial respiratory chain (37, 38). A series of experiments carried out in our laboratory to evaluate those factors that are limiting for ethanol oxidation in hepatocytes prepared from fed and 24-hour starved rats is shown in Table I. The rate of ethanol oxidation was about 40% greater in hepatocytes from fed rats compared to hepatocytes from fasted rats. If oxygen consumption, that is, the ability of the respiratory chain to reoxidize reducing equivalents is rate limiting for ethanol oxidation, the addition of uncoupling agents, which stimulate oxygen uptake, should increase the rate of ethanol oxidation. FCCP (carbonyl cyanide *p*-trifluoromethoxyphenylhydrazone) is a potent uncoupling agent; oxygen consumption by the liver cells was increased by 30% to 50% in the presence of FCCP. In liver cells prepared from fed rats, 1 and 2 μ M FCCP stimulated the rate of ethanol oxidation (Table I). The stimulatory effect was lost at higher concentrations of FCCP and inhibition was observed at 5 μ M FCCP. Careful titration is therefore required to observe the stimulatory effect of FCCP. By contrast, in liver cells

TABLE I

Effect of Components of Substrate Shuttles and Uncoupling Agents on Ethanol Oxidation by Hepatocytes Isolated from Fed and Fasted Rats

Addition	Fed		Fasted	
	Ethanol Oxidation (μ mol/h/g wet wt)	Effect (%)	Ethanol Oxidation (μ mol/h/g wet wt)	Effect (%)
A. None	68.5 $\pm$ 10.7	—	47.4 $\pm$ 3.1	—
1 μ M FCCP	91.1 $\pm$ 10.8	+33	—	—
2 μ M FCCP	91.1 $\pm$ 8.2	+33	39.5 $\pm$ 6.1	-17
3 μ M FCCP	74.1 $\pm$ 9.6	+8	31.3 $\pm$ 5.0	-34
5 μ M FCCP	39.3 $\pm$ 5.7	-43	23.4 $\pm$ 2.1	-51
B. None	57.00 $\pm$ 4.82	—	41.25 $\pm$ 2.23	—
10 mM malate	74.57 $\pm$ 4.36	+31	65.25 $\pm$ 5.40	+58
10 mM aspartate	71.00 $\pm$ 5.78	+25	60.16 $\pm$ 6.16	+46
10 mM glutamate	75.75 $\pm$ 4.68	+33	65.50 $\pm$ 3.72	+59
10 mM α -glycerophosphate	51.28 $\pm$ 5.33	-10	52.00 $\pm$ 6.56	+26
C. 0.05 mM Dinitrophenol + malate	91.00 $\pm$ 7.13	+60	79.66 $\pm$ 6.11	+93
0.05 mM dinitrophenol + glutamate	116.60 $\pm$ 10.44	+105	97.33 $\pm$ 9.78	+136

prepared from fasted rats, ethanol oxidation was inhibited by concentrations of FCCP which were stimulatory with liver cells prepared from fed rats (Table I, experiment A). These experiments support the concept that in liver cells from fed rats, but not fasted rats, the rate of ethanol oxidation is primarily limited by the capacity of the respiratory chain to reoxidize reducing equivalents. Dinitrophenol, another uncoupling agent, has been shown to increase the rate of ethanol oxidation (35).

The addition of the components of the malate-aspartate shuttle (malate, aspartate or glutamate) or the α -glycerophosphate shuttle (α -glycerophosphate) to cells from fasted rats increased the rate of ethanol oxidation by 30% to 60% (Table I, experiment B). Combinations of several components (malate + aspartate + glutamate + α -glycerophosphate) were not more effective in stimulating ethanol oxidation than were the individual components. In hepatocytes from fed rats, the addition of the components of the malate-aspartate shuttle caused a lesser extent of stimulation of ethanol oxidation (25% to 33%) than found with liver cells from fasted rats; α -glycerophosphate was not stimulatory with preparations from fed rats (Table II,

experiment B). In view of the greater extent of stimulation of ethanol oxidation by substrate shuttle components in hepatocytes from fasted rats compared to fed rats, the transfer of reducing equivalents via substrate shuttles into the mitochondria is especially limiting for ethanol oxidation in the fasted state.

The above results suggest that under conditions of sufficient intracellular levels of intermediates of the substrate shuttles, ethanol oxidation is regulated by the capacity of the respiratory chain to reoxidize reducing equivalents generated by the alcohol dehydrogenase reaction. This can be further seen from the results of experiment C (Table I). In hepatocytes from fed rats, the addition of malate or glutamate increased the rate of ethanol oxidation by 31% and 33%, respectively. However, in the presence of malate or glutamate plus the uncoupling agent dinitrophenol, the rate of ethanol oxidation was increased by 60% and 105%, respectively. In hepatocytes from fasted rats, the addition of malate or glutamate increased the rate of ethanol oxidation by 58% and 59%, respectively. In the presence of malate or glutamate plus dinitrophenol, the rate of ethanol oxidation was increased by 93% and 136%, respectively. Consequently, in the presence of substrate shuttle metabolites, uncoupling agents are effective in stimulating the rate of ethanol oxidation beyond the stimulation produced by the substrate shuttle components themselves. In summary, the concentration of substrate shuttle intermediates is rate limiting for the oxidation of ethanol by liver cells from fasted rats, whereas in liver cells from fed rats, the rate of ethanol oxidation is regulated by mitochondrial reoxidation of reducing equivalents.

It is important to note that none of the above conditions (pyruvate, shuttle metabolites, uncoupling agents or metabolites plus uncouplers) increased the rate of ethanol oxidation to a higher rate than the reported *in vivo* rate (about 200 μ mol/h/g, wet wt). Consequently, under certain conditions (maximal metabolite levels and oxygen consumption rates), conditions more associated with *in vivo* than *in vitro* conditions, ethanol oxidation may also be limited by the maximum capacity of alcohol dehydrogenase (40, 41).

The Microsomal Alcohol Oxidizing Pathway

In 1965, Orme-Johnson and Ziegler described a microsomal system that was capable of oxidizing methanol and ethanol (42). The subsequent detailed studies by Lieber and coworkers led to the full characterization of the microsomal ethanol (or alcohol) oxidizing system (MEOS or MAOS).

TABLE II

Effect of Chronic Ethanol Consumption on Ethanol Metabolism, Alcohol Dehydrogenase, Mitochondrial Activity, Shuttle Enzymes and Reconstituted Shuttle Activities

Reaction	Specific Activity		Effect of Ethanol (%)
	Control	Ethanol-treated	
Blood Ethanol Clearance (μ mol/min/kg body wt)	90 $\pm$ 9	130 $\pm$ 17	+44
Alcohol Dehydrogenase (nmol/min/mg soluble protein)	13.8 $\pm$ 1.5	11 $\pm$ 1	-20
Cytochrome Oxidase (K^{-1} /g liver)	929 $\pm$ 119	725 $\pm$ 100	-22
Cytochrome Oxidase (K^{-1} /mg mitochondrial protein)	14.2 $\pm$ 1.8	10.9 $\pm$ 1.5	-23
Mitochondrial Content (mg/g liver)	65.8	66.7	+1
Mitochondrial Aspartate Amino-transferase (nmol/min/mg mitochondrial protein)	1141 $\pm$ 66	1156 $\pm$ 106	+1
Cytosolic Aspartate Amino-transferase (nmol/min/mg soluble protein)	114 $\pm$ 6	118 $\pm$ 4	+4
Mitochondrial α -Glycerophosphate Oxidase (natom/min/mg mitochondrial protein)	9.8 $\pm$ 1.2	7.4 $\pm$ 1	-25
Cytosolic α -Glycerophosphate Dehydrogenase (nmol/min/mg soluble protein)	190 $\pm$ 20	150 $\pm$ 20	-21
NADH Oxidation (nmol/min/mg mito protein)	3.91 $\pm$ 0.63	3.60 $\pm$ 0.58	-8
Reconstituted Fatty Acid Shuttle (nmol/min/mg mito protein)	11.2 $\pm$ 1.3	11.1 $\pm$ 1.4	-1
Reconstituted α -Glycerophosphate Shuttle (nmol/min/mg mito protein)	16 $\pm$ 3.2	14.1 $\pm$ 1.4	-12
Reconstituted Malate-Aspartate Shuttle (nmol/min/mg mito protein)	17.1 $\pm$ 1.6	18.5 $\pm$ 2.1	+8

Chronic consumption of ethanol by rats resulted in proliferation of the smooth endoplasmic reticulum (43) and increased activity of a variety of hepatic microsomal drug metabolizing enzymes (44, 45) as well as an increase in cytochrome P-450 (45, 46) and NADPH oxidase (47). These effects of ethanol are similar to those of drugs that are metabolized by the microsomal cytochrome P-450-dependent mixed function oxidase system. The dependence of MEOS on NADPH, the partial inhibition by carbonmonoxide (which reacts with cytochrome P-450), and the ability of ethanol to competitively inhibit the metabolism of typical substrates (drugs) for the mixed function oxidase led to the hypothesis that the MEOS pathway involved components of the mixed function oxidase system (48, 49). Others ascribed the MEOS pathway to contamination of isolated microsomes by alcohol dehydrogenase or by catalase (50, 51). Differentiation of MEOS from alcohol dehydrogenase was based on differences in K_m values for ethanol, cofactor requirements, pH optimum and effects of pyrazole, an inhibitor of alcohol dehydrogenase (48). However, differentiation of MEOS from catalase has been a subject of great controversy. Microsomes can generate H_2O_2 from added NADPH, and ethanol is a known peroxidatic substrate for catalase. Hence, it is possible that MEOS may represent a combination of the activities of NADPH oxidase and catalase (present as a contaminant in isolated microsomal preparations) (50–52). Experiments involving inhibitors of catalase, measurements of the total capacity of H_2O_2 production by the liver, alcohols that are substrates for MAOS but do not serve as substrates for the peroxidatic activity of catalase, for example, 1-butanol, and physical separation of MEOS from catalase and alcohol dehydrogenase by column chromatography purification procedures (53, 54), appear to provide evidence to distinguish MEOS from catalase and establish the MEOS pathway as a separate minor pathway for ethanol metabolism by the liver. No MEOS activity was detected in microsome preparations from brain or heart muscle (48). A major advance in this area was the demonstration that reconstituted systems containing purified preparations of cytochrome P-450 were capable of oxidizing ethanol to acetaldehyde (55–58). These preparations are devoid of catalase and alcohol dehydrogenase activities.

Significance of MEOS

The main pathway for ethanol metabolism involves alcohol dehydrogenase. However, ethanol metabolism was found to persist even in the pres-

ence of pyrazole, a potent inhibitor of alcohol dehydrogenase (59–61). The extent of contribution of this non-alcohol dehydrogenase pathway to the total ethanol metabolism varied according to the concentration of ethanol used; from 10%–25% (48, 49) to half or more (59, 60) of the total ethanol metabolism. The rate of ethanol metabolism increases at higher concentrations of ethanol (59, 60). Alcohol dehydrogenase has a K_m for ethanol of about 1–2 mM whereas the K_m for ethanol for MEOS is about 10 mM. In view of the low K_m of alcohol dehydrogenase for ethanol, the increase in the rate of ethanol metabolism at elevated concentrations of ethanol appears to reflect the presence of a nonalcohol dehydrogenase pathway capable of oxidizing ethanol, presumably MEOS. In fact, in contrast to a long held belief that the kinetics of ethanol elimination from the blood followed zero order kinetics, recent studies reveal a distinct break in the kinetic curve for ethanol elimination (62). The kinetics of ethanol elimination were consistent with the presence of two pathways, one with a K_m for ethanol of about 8–10 mM, the other with a K_m for ethanol of about 1–2 mM (62). The former pathway has the same K_m for ethanol as the MEOS pathway. Additional studies involving the stereospecific labeling of glucose and H_2O from (1R- ^3H) ethanol (63) and the lack of significant effects of high concentrations of ethanol on the cytosolic redox state (64) also led to the conclusion of an alcohol dehydrogenase-independent pathway of ethanol metabolism, which contributes, in part, to the overall metabolism of ethanol.

Aside from playing an increasingly important role in ethanol metabolism at higher concentrations of ethanol, MEOS was found to increase in activity after chronic ethanol consumption (48, 49). Isolated microsomes from chronic ethanol-fed rats displayed about a two-fold higher rate of ethanol oxidation than microsomes from pair-fed controls (48, 49). Chronic administration of ethanol results in an increased rate of ethanol metabolism in vivo and in vitro (48, 49, 65–67). It has been suggested that MEOS can account for about two-thirds of the adaptive increase in ethanol oxidation found after chronic ethanol administration (48, 49). Thus MEOS appears to contribute to a greater extent to ethanol metabolism after chronic ethanol consumption.

After chronic ethanol consumption (but in the absence of ethanol), the metabolism of several drugs was found to be increased (Reviewed in 68). However, in the presence of ethanol, microsomal metabolism of drugs was depressed (68). These dual ef-

fects of ethanol on microsomal drug metabolism (stimulation after chronic consumption, inhibition in the presence of ethanol) have been ascribed to the ability of ethanol to be oxidized by microsomes, the MEOS pathway (68). In the presence of ethanol, drug metabolism is depressed because ethanol competes with drugs for oxidation by the cytochrome P-450 mediated mixed function oxidase pathway. After chronic ethanol consumption, proliferation of the endoplasmic reticulum occurs, MEOS is induced, and there is an increased content of cytochrome P-450 and other microsomal electron transfer components (48, 68). As a result the oxidation of other substrates i.e., drugs, is increased as long as ethanol is not present to compete with the drugs for the microsomal mixed function oxidase system. These interactions of ethanol with microsomes may play an important role in the increased sensitivity of "drinkers" to certain drugs, as well as the resistance of "alcoholics" to drugs (68).

Ethanol Metabolism after Chronic Ethanol Consumption

Chronic administration of ethanol increases the rate of ethanol metabolism (48, 49, 65-67). The mechanisms underlying this increase are controversial. With regard to the effects of chronic ethanol consumption on the activity of enzyme systems which oxidize ethanol to acetaldehyde, alcohol dehydrogenase activity has been reported to be increased (19, 65), not changed (35), or even slightly decreased (47), after chronic ethanol intake. As mentioned above, MEOS activity adaptively increases after chronic ethanol consumption. In view of altered mitochondrial structure as evidenced by ultrastructural examination (69) and reports of increased membrane permeability (70), it has been suggested that the normal permeability barrier to NADH had been altered by the ethanol treatment and that transport of substrate metabolites into and out of the mitochondria may be augmented (71, 72). Therefore, enhanced NADH permeability and shuttle activities would contribute to the enhanced rate of ethanol metabolism. Since about 60% of the oxygen consumption in the liver is utilized to reoxidize NADH formed from the oxidation of ethanol (36), increased mitochondrial activity or content per g of tissue may play a role in the adaptive increase in ethanol metabolism. In this regard, a major hypothesis to account for the metabolic increase in ethanol metabolism was advanced by Israel and collaborators (67, 73, 74). Chronic ethanol consumption was suggested to result in the production of a hypermetabolic state of the liver, thereby

accelerating the reoxidation of NADH produced by the alcohol dehydrogenase pathway. The hypermetabolic state was hypothesized to result from an increase in $(\text{Na}^+ + \text{K}^+)$ -activated ATPase, the increase in generation of ADP bringing the hepatocyte closer to "state 3", thereby increasing mitochondrial oxygen consumption. This ethanol-induced hypermetabolic state may be mediated by thyroid hormones and possibly catecholamines. In fact, the antithyroid drug propylthiouracil has been used to prevent liver cell damage produced by ethanol (75).

To evaluate some of the above possibilities (76-78), mitochondria were isolated from male Sprague-Dawley rats that were fed a liquid diet for about four weeks and in which ethanol provided 36% of total calories, protein 16%, fat 35% and carbohydrate 13%. Pair-fed littermates were given a similar diet except that carbohydrate replaced ethanol isocalorically (79). This diet, developed by DeCarli and Lieber, has been shown to result in the production of a fatty liver (79). As shown in Table II, the rate of blood ethanol clearance was increased by 44% in rats fed ethanol chronically. However, the activity of alcohol dehydrogenase was decreased 20%, indicating the increased rate of ethanol metabolism is not mediated by a parallel increase in the activity of this enzyme. Mitochondria isolated from ethanol-fed rats displayed lower rates of oxygen consumption with a variety of substrates compared to mitochondria from pair-fed controls (77, 78). The activity of cytochrome oxidase was lower in homogenates and in isolated mitochondria from ethanol-fed rats (Table II). Total mitochondrial protein per g of liver (as calculated from the total and specific activities of cytochrome oxidase) was not altered by chronic ethanol consumption. Similar results were obtained with another marker mitochondrial enzyme, succinic dehydrogenase (76). The above results, and other assays of mitochondrial functions (77, 78, 80), revealed that chronic ethanol consumption decreased, rather than increased, isolated mitochondrial activity. The activity of enzymes participating in the malate-aspartate or α -glycerophosphate shuttles were either not affected, or slightly decreased by chronic ethanol feeding (Table II). When the shuttles were reconstituted with mitochondria from ethanol-fed rats or from pair-fed controls, identical rates of NADH oxidation were observed (Table II). Further studies on mitochondrial permeability indicated that various inhibitors of anion transport across the mitochondrial membrane e.g. iodobenzyldmalonate, avenaciolide, mersalyl, were effective in preventing malate-aspartate

shuttle activity to comparable extents with mitochondria from ethanol-fed rats and controls (76). Moreover, swelling studies in isotonic solutions of ammonium salts (an assay of the penetration of anions into the mitochondria, which are freely permeable to NH_3) indicated that mitochondria from control rats swelled to the same extent as those from ethanol-fed rats when suspended in the ammonium salts of phosphate, malate, succinate, glutamate or citrate (76). Thus, chronic ethanol consumption produced no change in the permeability of the mitochondria to these anions. In addition, the normally low permeability of intact mitochondria to NADH was maintained by mitochondria from ethanol-fed rats (Table II). In summary, the increased rate of ethanol metabolism found after chronic ethanol consumption does not correlate with the activity of alcohol dehydrogenase, *in vitro* mitochondrial activity, content of mitochondria, enhanced shuttle activity or permeability to various substrate shuttle anions and NADH.

Since various results obtained with isolated mitochondria did not correlate with the increased rate of ethanol oxidation, studies with isolated liver cells prepared from the ethanol-treated rats were undertaken (81). The rate of ethanol oxidation in liver cells isolated from ethanol-fed rats was $78 \mu\text{mol/h/g}$ liver, wet wt, compared to a value of $56 \mu\text{mol/h/g}$ liver for controls (Table III). The increase of 39% in the rate of ethanol oxidation is comparable to that found *in vivo* (Table II), indicating that the increase in ethanol metabolism involves pathways associated with the liver. Methylpyrazole, a potent inhibitor of alcohol dehydrogenase, decreased the rate of ethanol metabolism by 86% in liver cells isolated from controls, and by 75% in liver cells from ethanol-fed rats (Table III). These results confirm the dominant role of the alcohol dehydrogenase pathway in both liver cell preparations. However, the results also suggest that a non-alcohol dehydrogenase-dependent pathway contributes a larger fraction to the overall pathway of ethanol metabolism in the liver cells from the ethanol-fed rats. Indeed, about one-half of the increase in the rate of ethanol metabolism persisted in the presence of concentrations of methylpyrazole which totally block the activity of alcohol dehydrogenase. Similar results were obtained with other inhibitors of the alcohol dehydrogenase pathway, that is, about 40% to 50% of the increase in the rate of ethanol oxidation was not abolished by inhibitors of the respiratory chain (rotenone, antimycin) inhibitors of ADP-dependent oxygen consumption (oligomycin, atracyloside) and inhibitors of substrate shuttle anion

TABLE III
Rate of Ethanol Metabolism in Liver Cells Isolated from Chronic Ethanol-Fed Rats and Pair-Fed Controls

Addition	Rate of Ethanol Oxidation		In-crease in Ethanol Oxida-tion
	Control	Chronic Ethanol	
	(μmol/h/g liver, wet wt)		
A. None	56.5 ± 3.6	78.4 ± 3.9	21.9
Methylpyrazole	8.0 ± 2.3	17.5 ± 1.5	9.5
Rotenone	32.7 ± 3.8	44.0 ± 14.3	11.3
Antimycin	14.4 ± 3.5	23.8 ± 2.3	9.4
Oligomycin	30.5 ± 3.6	39.5 ± 2.8	9.4
Atractyloside	33.0 ± 4.8	45.6 ± 4.1	12.6
Amino-Oxyacetate	32.5 ± 4.3	43.9 ± 3.9	11.4
Avenaciolide	32.8 ± 3.9	44.8 ± 3.4	12.0
Iodobenzylmalonate	26.2 ± 5.6	36.5 ± 3.1	10.3
Ouabain	42.5 ± 4.6	62.9 ± 5.5	20.4
B. None	54.0 ± 3.7	69.6 ± 5.4	15.6
Malate	60.1 ± 5.7	80.2 ± 9.8	20.1
Aspartate	60.3 ± 3.6	83.3 ± 6.4	23.0
Glutamate	60.0 ± 3.9	74.3 ± 5.6	14.3
α-Glycerophosphate	57.7 ± 4.3	71.1 ± 5.3	13.4
Aspartate + Dinitro-phenol	72.9 ± 3.6	91.1 ± 5.2	18.2
Glutamate + Dinitro-phenol	103.7 ± 5.8	124.0 ± 9.1	20.3
α-Glycerophosphate + Dinitrophenol	71.2 ± 6.2	88.7 ± 7.9	17.5

transport (amino-oxyacetate, avenaciolide, iodobenzylmalonate) (Table III). Furthermore, the entire increase in the rate of ethanol oxidation persisted in the presence of various substrate shuttle metabolites, in the absence or presence of an uncoupling agent (dinitrophenol), suggesting that the increased rate did not reflect differences in hepatic content of these metabolites (Table III).

There was no effect by ouabain, an inhibitor of the ($\text{Na}^+ + \text{K}^+$)-activated ATPase, on the increased rate of ethanol oxidation (Table III), suggesting that increased activity of this enzyme, with the subsequent development of a state 3 condition did not occur. Indeed, hepatic oxygen consumption with a variety of substrates, or in the presence of dinitrophenol, was slightly decreased, not increased, after chronic ethanol consumption (Table IV). These results are, in general, not consistent with the development of a hypermetabolic state after chronic ethanol consumption. In view of the demonstrated increase in MEOS activity after chronic ethanol consumption (48, 49), the greater extent of increase in ethanol metabolism at higher ethanol concentrations (82), the inability of pyrazole and methylpyrazole to suppress the increased rate of ethanol metabolism (49, 81, 82), and the persistence of at least part of the increased rate of ethanol

metabolism in the presence of various inhibitors of the alcohol dehydrogenase pathway (Table III), pathways other than alcohol dehydrogenase appear to play a role in the metabolic adaptation which occurs after chronic ethanol consumption.

Recent Studies on the Nature of MEOS

The detailed investigations of Lieber and coworkers demonstrated that MEOS activity can be separated from the peroxidatic activity of catalase and that MEOS may play an important role in the metabolic adaptation found after chronic ethanol consumption. An important development was the finding that purified preparations of cytochrome P-450 could oxidize ethanol in the presence of NADPH, phospholipid and NADPH-cytochrome c reductase (55-58). Chronic ethanol consumption resulted in an increase of a cytochrome P-450 he-

moprotein which was distinct from that induced by drugs such as phenobarbital or 3-methylcholanthrene (57). This purified cytochrome P-450 was more active for alcohol oxidation than control cytochrome P-450 preparations (57). The mechanism responsible for the oxidation of ethanol and other alcohols by microsomes at the molecular level remains to be established. Our laboratory has recently investigated the possibility that MEOS may represent the interaction of ethanol with hydroxyl radicals generated from microsomal electron transfer (83-86). There is substantial evidence that ethanol is an effective scavenger of hydroxyl radicals in vitro and in vivo; the product of the reaction being acetaldehyde (Reviewed in 87). The addition of a series of compounds that are known to react with hydroxyl radicals ($\cdot\text{OH}$) on the activity of MEOS was therefore evaluated. Thiourea, dimethylsulfoxide (DMSO) mannitol and benzoate effectively inhibited the activity of MEOS (Fig. 2). Corresponding controls, which are not scavengers of $\cdot\text{OH}$, for example, urea, had no effect on MEOS. Kinetic experiments indicated that the $\cdot\text{OH}$ scavengers were competitive inhibitors of MEOS, that is, they competed with ethanol for $\cdot\text{OH}$. The inhibition by the $\cdot\text{OH}$ scavengers was specific since these agents had no effect on catalase-dependent oxidation of ethanol, on microsomal drug metabolism (aminopyrine demethylation, aniline hydroxylation) or on microsomal electron transfer (83, 84). These results dissociate the overall pathways of MEOS from drug metabolism.

TABLE IV

Rate of Oxygen Consumption in Liver Cells Isolated from Chronic Ethanol-fed Rats and Pair-fed Controls

Addition	Rate of Oxygen Consumption	
	Control	Chronic Ethanol
	($\mu\text{mol}/\text{min}/\text{mg}$ liver cell protein)	
None	2.45 ± 0.13	2.22 ± 0.21
Glutamate	2.79 ± 0.17	2.53 ± 0.23
Pyruvate	2.49 ± 0.09	2.19 ± 0.41
α -Glycerophosphate	2.88 ± 0.18	2.66 ± 0.22
Succinate	4.35 ± 0.37	4.02 ± 0.44
Dinitrophenol	3.36 ± 0.28	3.01 ± 0.39

EFFECT OF THIOUREA, DMSO, MANNITOL AND BENZOATE ON MICROSOMAL OXIDATION, OF ETHANOL IN THE PRESENCE OF AZIDE

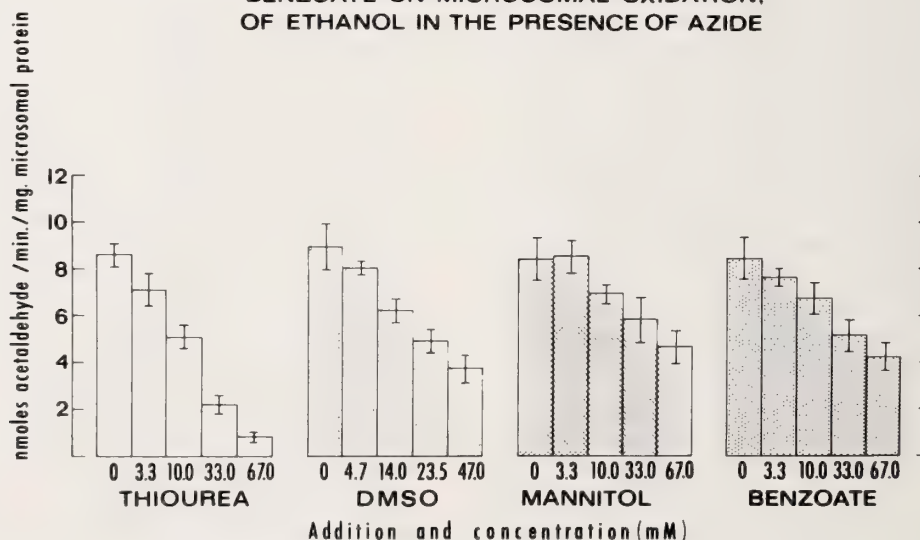


FIG. 2. Effect of thiourea, dimethylsulfoxide (DMSO), mannitol and benzoate on microsomal oxidation of 51 mM ethanol. Experiments were carried out in the presence of 0.5 mM azide.

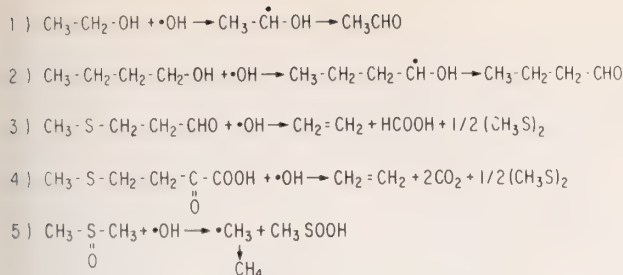


FIG. 3. Chemical equations for the interaction of hydroxyl radical scavenging agents with $\cdot\text{OH}$. The equations represent the interaction of ethanol (1), 1-butanol (2) methional (3) KTBA (4) and DMSO (5) with $\cdot\text{OH}$ to produce acetaldehyde (1) butyraldehyde (2), ethylene (3 and 4) and methane (5).

Do Microsomes Generate ·OH

Fig. 3 shows chemical equations for the interaction of various $\cdot\text{OH}$ scavenging agents, namely ethanol, 1-butanol, methional, 2-keto-4-thiomethylbutyric acid (KTBA) and DMSO with $\cdot\text{OH}$ to produce known products, acetaldehyde, butyraldehyde, ethylene and methane. Rat liver microsomes incubated with an NADPH-generating system produced acetaldehyde from ethanol, butyraldehyde from 1-butanol, ethylene gas from methional and KTBA, and methane gas from DMSO (85, 86). Cross competition experiments indicated that when ethanol (or butanol) was present, ethylene and methane generation were decreased, whereas in the presence of methional, KTBA and DMSO, acetaldehyde production from ethanol was decreased (Table V). These results are consistent with the ability of the five agents listed in Fig. 3 to compete for $\cdot\text{OH}$ generated by microsomes.

H₂O₂ as a Precursor of ·OH

Two H_2O_2 -dependent pathways which may give rise to $\cdot\text{OH}$ are a) the Fenton reaction (88) $\text{Fe}^{2+} + \text{H}_2\text{O}_2 \rightarrow \text{Fe}^{3+} + \cdot\text{OH} + \text{OH}^-$ b) the Haber-Weiss reaction (89) $\text{O}_2^{\cdot-} + \text{H}_2\text{O}_2 \rightarrow \text{O}_2 + \text{OH}^- + \cdot\text{OH}$. Microsomes generate $\text{O}_2^{\cdot-}$ (superoxide anion) and H_2O_2 during NADPH oxidation and thus have the potential to generate $\cdot\text{OH}$. Azide, which is a potent inhibitor of catalase, stimulated the production of ethylene from methional and KTBA, of methane from DMSO and of butyraldehyde from 1-butanol (Table VI). Azide, by inhibiting catalase, promotes the accumulation of H_2O_2 , a precursor of $\cdot\text{OH}$. Increased generation of $\cdot\text{OH}$ would result in increased generation of ethylene, methane and butyraldehyde. Additional experiments indicated that the external addition of H_2O_2 to microsomes resulted in an increase in NADPH-dependent oxida-

TABLE V
*Competition Between Ethanol and Other Hydroxyl Radical
Scavengers*

Substrate	Competitor	Rate of Reaction	
		−Competitor	+Competitor
10 mM Methional	33 mM Ethanol	27.7 ± 1.1	15.0 ± 1.3
10 mM KTBA	33 mM Ethanol	28.3 ± 0.8	22.1 ± 1.2
33 mM DMSO	33 mM Ethanol	2.5 ± 0.3	1.6 ± 0.3
33 mM Ethanol	10 mM Methional	249 ± 14	131 ± 9
33 mM Ethanol	10 mM KTBA	270 ± 16	131 ± 9
33 mM Ethanol	33 mM DMSO	233 ± 15	80 ± 5

TABLE VI
*Effect of Azide on Microsomal Metabolism of Hydroxyl
Radical Scavenging Agents*

Scavenger	Product	Rate of Reaction	
		−Azide	+Azide
		(nmol/19min/mg protein)	
Methional	Ethylene	11.4 ± 1.7	29.0 ± 1.4
KTBA	Ethylene	11.0 ± 1.9	25.0 ± 1.8
DMSO	Methane	0.55 ± 0.31	1.73 ± 0.41
1-Butanol	Butyraldehyde	72.2 ± 3.6	168.6 ± 9.7

TABLE VII
Effect of ·OH Scavengers on Ethanol Oxidation and Drug Metabolism by Purified Cytochrome P-450

Scavenger	Turnover Number		
	Ethanol Oxidation	Benzphetamine Demethylase	Aminopyrine Demethylase
	(nmol/min/nmol Cytochrome P-450)		
Control	27.7	22.1	11.7
DMSO			
5 mM	21.7	20.1	—
15 mM	15.4	20.8	13.4
30 mM	10.9	20.8	15.1
60 mM	6.3	20.5	—

tion of ethanol and 1-butanol (83). A consistent interpretation of these data is that H_2O_2 serves as a precursor of $\cdot\text{OH}$.

Ethanol Oxidation by Purified Cytochrome P-450

Ethanol oxidation by a reconstituted system composed of cytochrome P-450 purified from liver microsomes of phenobarbital-treated rats, NADPH-cytochrome C reductase, phospholipid and NADPH was inhibited by a series of $\cdot\text{OH}$ scavengers (90). Table VII shows a typical experiment utilizing DMSO as the $\cdot\text{OH}$ scavenging agent. Inhibition was competitive with respect to ethanol and was specific since drug metabolism was not affected (Table VII). The purified system generated ethylene from KTBA thereby providing chemical evidence consistent with the ability of purified cytochrome P-450 to generate $\cdot\text{OH}$ (90).

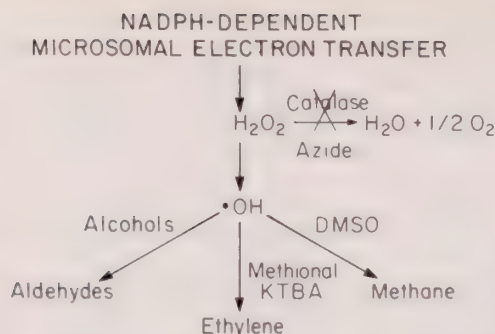


FIG. 4. Scheme for the generation of $\cdot\text{OH}$ and interaction with various $\cdot\text{OH}$ scavenging agents. Azide, by inhibiting catalase, allows H_2O_2 to accumulate. The increased accumulation of H_2O_2 results in increased generation of $\cdot\text{OH}$, and subsequently of ethylene, methane and butyraldehyde.

Summary

In summary, the ability of known $\cdot\text{OH}$ scavenging agents to competitively inhibit microsomal and purified cytochrome P-450-dependent oxidation of ethanol, without affecting associated microsomal reactions such as drug metabolism, suggests that MEOS involves the interaction of alcohols with $\cdot\text{OH}$ generated by microsomal electron transport. Chemical evidence consistent with the ability of microsomes to generate $\cdot\text{OH}$ is provided by the studies measuring generation of the hydrocarbon gases ethylene and methane. Experiments with azide and H_2O_2 indicate that H_2O_2 may serve as a precursor of $\cdot\text{OH}$. A scheme summarizing these results is shown in Figure 4. Studies concerning the generation of $\cdot\text{OH}$ after chronic ethanol consumption are in progress. In this regard, increased generation of $\cdot\text{OH}$ after chronic ethanol consumption may contribute to the increase in MEOS activity, to the metabolic adaptation found after chronic ethanol consumption, and possibly to the toxic actions of ethanol to the liver cell.

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Student's Corner

Paul Celano, BS, Editor

Air Pollution and Its Effects on Adult Asthmatics: A Review

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Introduction

In recent years, research on the relationship between environmental exposures to certain air pollutants and the exacerbation of asthma has intensified. The purpose of this paper is to present an overview of the important research findings to date, both experimental and epidemiologic.

Despite the increase in research, our understanding of the effects of common air pollutants upon the asthmatic is still in its infancy, partially because of the myriad of conflicting results investigators have reported. The results differ because air pollutants differ both in type and concentration in different locations, and at different times, and also because their effects on a biological system may depend upon complex interrelationships with other air pollutants (both known and currently unknown pollutants) as well as with meteorologic factors (1). In addition, the individual's actual exposure is influenced by such factors as cigarette smoking (1-3), occupational environments, socioeconomic factors, and the amount of time spent indoors (3-6). Also, it is now known that there are other agents besides air pollution that are presently believed to have the capability of bringing about an asthmatic episode. For instance, a number of different illnesses, such as particular psychologic states (10-12), bronchitis (13-16), and allergy (17-18), predispose one to asthma (7-9). Additional insults or triggers (10)

that may induce an asthmatic attack include meteorologic factors (such as climatic extremes or changes (19-27)), exercise (28-40), emotional stress (12, 41-42), respiratory infection (13, 43-44), aspirin and 'aspirin-like' drugs (45-47), allergens, irritants (perfumes, some cosmetic sprays, tobacco smoke, cooking odors, petroleum solvents, household deodorizing sprays and many industrial dusts and fumes) (48-55), pollens, molds (56-60), spores, pain, and foods (14). Though air pollution is being stressed in this review, the importance of other factors that may have effects on the asthmatic should not be dismissed.

It must be emphasized that asthma, despite the simplicity implied by many investigators, is actually a complex disease with a multifactorial causation. Asthma can be briefly defined as a clinical syndrome characterized by partial, variable obstruction of the peripheral airways which is reversible with time, either spontaneously or as a result of therapy (61).

Air pollution, which is a pulmonary pathogen, commonly consists of mixed oxidant and irritant gases and particulate matter. For the purpose of this presentation, the definition of air pollution as stated in the Report of the Air Conservation Commission (62) will be used: air pollution occurs "when (man-made) wastes are produced so rapidly, or when they accumulate in such concentrations that the normal, self-cleansing or dispersive propensities of the atmosphere cannot cope with them." In this paper a discussion of radioactive substances has been omitted. The major air pollutants known at this time include carbon monoxide, metals, particulate matter, oxides of sulfur, and photochemical

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pollutants. The chemical and physical characteristics of these pollutants will not be dealt with here; good reviews of this information have been published in recent years (62-65).

Experimental Research

Ozone has been a major focus of recent research. Experimental data have shown that concentrations of oxidant pollutants that are frequently found in some areas of the United States can have marked effects on experimental animals and human subjects. Coffin et al (66) found that 0.15 ppm ozone for several hours would lower the resistance of mice against streptococcal aerosols, and Murphy et al (67) observed high levels of various oxidants in the air cause an increase in respiratory resistance and increases susceptibility to respiratory infection. Similarly, in Los Angeles, Swann et al (68) found guinea pigs developed increased airway resistance when exposed to smog. However, Stokinger (69) presented evidence that rats under physical exercise can develop an ozone tolerance with a cross tolerance for nitrogen dioxide by exposing the animals to an ozone concentration of 0.3 ppm. Slonim et al (70) maintain that inhalation of ozone (0.6-0.8 ppm) for two hours is required to elicit pulmonary flow rate reductions, while Goldsmith and Nadel (71) found that there are individuals who develop bronchoconstriction on exposure to 0.1 ppm of ozone. Young, Shaw, and Bates (72) have evidence that inhalation of 0.70 ppm of ozone by normal individuals markedly increases airway resistance, while higher concentrations causes bronchoconstriction. Hazucha et al (73) provided evidence that inhalation of air with 0.37 and 0.75 ppm of ozone caused respiratory function impairment in healthy human subjects. Bennet (74) examined two groups with six healthy subjects in each, who were exposed to 0.2 ppm and 0.5 ppm ozone respectively for three hours per day, six days a week for 12 weeks. Those exposed to 0.2 ppm ozone were apparently unaffected, while those exposed to 0.5 ppm ozone showed reversible signs of irritation of terminal bronchi and bronchioles. Furthermore, a number of other studies with animals and humans have presented data that the inhalation of ozone, sulfate, and particulate matter can initiate bronchoconstriction, especially in sensitive subjects (55, 71, 75). Linn et al (76) exposed 22 asthmatic subjects to two-hour controlled exposures of 0.2 ppm ozone, in addition to secondary stresses of heat and intermittent exercise.

No meaningful changes in lung volumes, forced expiratory measures or single-breath N_2 indices were observed, and an insignificant increase in symptoms were found. However, significant blood biochemical changes did occur, for there were increased glucose-6-phosphate dehydrogenase and lactate dehydrogenase activities, increased erythrocyte fragility, and decreased concentration of reduced glutathione. In addition, there is increasing evidence to support the contention that low-level ozone exposures give rise to significant biochemical and morphologic lung lesions (77-80).

As with ozone, several studies (81-83) have offered evidence that in humans and animals, there is a marked increase in airway resistance after inhalation of sulfur dioxide. Speizer et al (84-85), after long-term studies of human subjects, including asthmatics, correlated decreases from predicted vital capacity with increased levels of sulfur dioxide from 0.5 ppm. Similarly, nitrogen dioxide inhalation also has been found by investigators to cause an increase in airway resistance (86), among other deleterious results (87).

Epidemiologic Studies

In 1948, an extended period of thermal inversion occurred in Donora, Pennsylvania, an intensely industrialized town. The occurrence of greatly increased levels of various pollutants in the air, particularly sulfur dioxide, adversely affected nearly 100% of the asthmatics in the area, while only 40% of the general population was deleteriously affected (88). Similarly, it was observed in London that asthmatics suffered a higher incidence of morbidity and mortality than the general population in 1952 (89) as well as in 1962 (90) when severe air pollution episodes occurred.

As early as 1954, increased incidence of asthmatic symptomatic flare-ups of military personnel who were stationed on the Kanto Plains in the industrialized Tokyo-Yokohama area had been reported. Frequently, improvement of the patient's condition was obtained by removal from the highly polluted area, though occasionally pulmonary function impairment had been observed (91-96).

Since the late 1950s, it has been noticed that the city of New Orleans has had periodic outbreaks of obstructive pulmonary disease (97-99); however, no relationship was observed between chemical air pollutants and these outbreaks in studies performed by the United States Department of Health, Edu-

cation, and Welfare (100), and only atopic individuals of the population seem to have been affected (101).

Oxidizing pollution worsens the conditions of people with pulmonary disease (24, 102). In a three-month study of 157 asthmatics in the Los Angeles area, Schoettlin and Landau (24) noticed a low positive correlation between chemical measures of air pollution and the number of asthmatics having attacks; when oxidant levels were greater than 0.14 ppm, the number of asthmatic episodes of the subjects increased.

Winkelstein et al (103) found that the mortality rate more than doubled for white males 50 to 69 years of age for asthma, emphysema, and bronchitis in high pollution areas as compared to low pollution areas. Glasser, Greenberg, and Field (104), during an episode of high air pollution in New York City (November 23–25, 1966) examined mortality and morbidity patterns. They discovered there was a rise on the third day of the episode in the number of emergency room visits for bronchitis and asthma among patients 45 years of age and older at three of the seven large New York hospitals examined. Unfortunately, the data available did not enable an evaluation of the contribution of the Thanksgiving Day holiday period to the observed increase in the emergency room visits.

Zeidberg et al (105–107) after studying 84 adult asthmatics in Nashville concluded that the frequency of asthmatic attacks was significantly associated with air pollution, as measured by the level of sulfation, particularly sulfur dioxide. Carnow et al (108) presented evidence from a study in Chicago that high levels of sulfur dioxide precipitate and exacerbate respiratory symptoms in people with chronic pulmonary diseases, including asthma. Nmekata and Carnow (22) recently examined the admission rates for all respiratory diseases in fourteen hospitals in the Chicago area and found them to be not only consistent with the fluctuations of sulfur dioxide, but also to be correlated with the average temperature and average percentage of sunshine. It remains unclear, however, which factor influences these admission rates most directly, and the extent to which it influences the condition of the asthmatic. Ramsey (23) in his study of seven asthmatics, observed that although urban levels of certain air pollutants, including suspended particulate and ozone, frequently were over the present federal standards, such conditions did not substantially aggravate the asthmatic condition. Goldstein and Block (109) in a study of emergency room visits

for asthma in two inner city areas of New York City, found that in one such area a positive correlation between sulfur dioxide and the number of daily visits existed, while in the other region no such correlation was present. No correlation was found in either region with smokes shade. They postulated another causative agent, that is disseminated in a similar pattern to sulfur dioxide, was responsible for their results.

Lewis (97) in a study of emergency room visits by asthmatic patients in New Orleans, correlated the high incidence of visits of such people to inhalation of air polluted with a high level of poor combustion particles containing a siliceous material. On the other hand, Schoettlin and Landau (24) found no significant correlation between the number of asthmatic subjects affected and the air levels of carbon monoxide and particulate matter. In separate studies Cohen et al (110) and Lebowitz et al (27) found a positive correlation between the amount of air pollution and the increased frequency of asthma attacks.

Currently, hospitals in Los Angeles have reported the admission and outpatient treatment of a far greater number of people than normal for aggravation of pre-existing respiratory ailments during a September 1979 smog episode, the worst such occurrence since 1955 (C. Inouye, personal communication). An investigation of this episode will undoubtedly yield valuable information regarding the effects of air pollution upon asthmatics.

Conclusion

Currently, attempts at explaining research observations on air pollution and its relationship to asthma are fraught with difficulties, chiefly because our ignorance on this subject greatly exceeds our knowledge. The amount of experimental research conducted to date is very limited, probably for ethical and logistic reasons. Our only hope in understanding asthma and the effects of air pollution upon the asthmatic, as well as its effects on those with any acute or chronic respiratory disease, rests in prophylactic research. Many, if not all, of the factors that can provoke an asthmatic attack will be found to work through a common mechanism of action. Air pollution is one of the more intricate triggers, because it is dependent upon numerous complex variables, many of which, like a number of the air pollutants themselves, are presently unknown. If most triggers of asthmatic attacks work

by a common mechanism, then it is vital to support research regarding both the effects of air pollution and the effects of other influences upon asthma. Only in this way can a true understanding of the asthmatic condition be achieved, and the patient thereby helped.

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Evaluation of Corticosteroid Treatment in Aspiration of Gastric Contents: A Controlled Clinical Trial

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Abstract

A double-blind controlled, clinical trial of treatment of aspiration of gastric contents in 60 patients was undertaken to evaluate the efficacy of corticosteroids. The patients studied could be divided into two subgroups, a younger group with drug overdose and an older group with predominantly neurologic disorders. Corticosteroid treatment in the overdose group reduced normalization time of arterial blood gas studies by 33 percent, x-ray normalization time by 45 percent, duration of intubation and mechanical ventilation by 55 percent and 49 percent, and length of stay in the intensive care unit by 26 percent. Although corticosteroid treatment reduced the time for x-ray and arterial blood gas normalization in the neurologic group, patients remained in the intensive care unit longer. There were no significant differences in the occurrence of complications or of mortality between treatment and control groups. This study indicates that corticosteroids hasten recovery in young drug overdose patients with aspiration of gastric contents but do not affect mortality or incidence of complications.

Aspiration of gastric contents results in pulmonary injury due to acidity of gastric juice (1, 2). After Mendelson's original description of pulmonary aspiration in obstetric patients, several studies stressed the importance of pH, volume and distribution and presence or absence of particulate matter and bacteria in gastric contents in determining the clinical outcome (3-7). Management of this syndrome has primarily been supportive. Corticosteroids have been used since 1955 with varying results (2, 8-13). Recent experimental studies (4, 14-16) and clinical observations (17) have indicated a lack of effectiveness of corticosteroids in treatment of pulmonary aspiration.

The purpose of this double-blind, controlled clinical trial was to study the efficacy of corticosteroids in treatment of aspiration of gastric contents. We believe this is the first double-blind controlled clinical trial reported in pulmonary aspiration.

Materials and Methods

A double-blind, controlled clinical trial was undertaken in 60 patients, using methylprednisolone sodium succinate (Solumedrol®) or a placebo. Size of the study was made sufficiently large to detect a one-day difference in mean x-ray and arterial blood gas normalization times between treatment and control groups with alpha and beta errors of 0.05 and 0.10 respectively.

The diagnosis of aspiration of gastric contents was made using the following criteria: the presence of a predisposing factor such as altered consciousness, either history or observation of vomiting and removal of gastric contents from the tracheobronchial tree, and the presence of x-ray and arterial blood gas abnormalities. Forty patients had gastric material removed from their endotracheal tubes. Of the remaining patients, eight were observed to aspirate in the emergency room and 12 patients had very convincing histories of aspiration given by their families.

All patients were admitted to the respiratory care unit or the medical intensive care unit at Mount Sinai Services-City Hospital Center at Elmhurst. Patients with active gastrointestinal bleeding or recent history of peptic ulcer, known heart disease

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seen as congestive heart failure, other known preexisting lung disease, pulmonary contusions, neurogenic pulmonary edema, and those who were diagnosed 24 hours or more after pulmonary aspiration were excluded from the study. Informed consent was obtained from patients or family of comatose patients.

Gastric pH, arterial blood gas determinations, chest x-rays, complete blood counts, serum electrolytes, stool guaiac examinations, and sputum cultures were obtained on admission and daily. The initial, worst in first 24 hours, and daily x-rays until normalization were reviewed by the authors. The day of x-ray normalization was determined when complete resolution of parenchymal infiltrates was noted.

The arterial/Alveolar ratios were calculated on the initial, worst in first 24 hours, and daily arterial blood gases until normalization (a/A ratio > 0.8) by using the simplified alveolar air equation:

$$P_{A}O_2 = P_{I}O_2 - PaCO_2 \times \frac{1}{R}$$

with R , the respiratory exchange ratio, assumed to equal 0.8 (18).

All the patients received standardized treatment as follows: oxygenation with or without assisted ventilation and positive end expiratory pressure, vigorous suctioning, careful administration of intravenous fluids, antacids every four hours for three or more days, crystalline penicillin 2.4 million units per day intravenously for community acquired disease (aspiration outside of hospital or within 12 hours of hospitalization) or clindamycin and gentamicin in usual doses for hospital acquired disease for a period of seven to ten days. Positive end expiratory pressure was given to patients with refractory hypoxia ($PO_2 < 50$ mm Hg on $0.5 F_{I}O_2$) until improvement in the arterial oxygen tension. Patients were extubated when clinical conditions improved according to standard bedside criteria for extubation (19).

The use of corticosteroids or placebo was randomized by the supplier (Upjohn) and given sequentially. Patients received either methylprednisolone sodium succinate 15 mg/kg body weight daily in three divided doses intravenously for three days or a placebo in a strict double-blind manner.

Complications were promptly recognized and treated. Major gastrointestinal bleeding was diagnosed when a fall in hematocrit of five percent or more occurred along with blood in nasogastric tube aspirates or positive stool guaiac tests. Superimposed bacterial pneumonia was diagnosed from new

x-ray changes after 48 hours consistent with pneumonia, worsening of arterial blood gas studies after 48 hours, increase in the white blood cell counts and fever after 48 hours, and repeatedly culturing a specific organism in sputum and/or blood.

Patients were discharged from the intensive care unit (ICU) when they no longer required intensive care. None of the patients remained in the ICU after recovery from aspiration pneumonitis and complications of the disease or treatment. After discharge from the ICU, patients were closely followed in the medical wards or chest clinic. Pulmonary function tests were done on available patients to evaluate the long-term sequelae of pulmonary aspiration and results will be reported in the future.

Fischer's exact test and analysis of variance was used to evaluate the data in this study. Length of stay data were analyzed after transformation to a log normal distribution.

Results

The mean age of patients was 40.6 years. There were 39 men and 21 women. Thirty-one patients received steroids and 29 patients received placebo.

In order to check for proper randomization, associations of steroid treatment with age, predisposing diagnosis, source of patients (community or hospital acquired aspiration), admission x-rays, arterial blood gases, gastric pH, intubation with or without mechanical ventilation, and presence of hypotension were evaluated. None of these associations was statistically significant ($P > .6$).

There was a significant association between predisposing diagnosis and sex and age (Table I). The majority of patients admitted for drug overdose (includes nine patients with narcotic overdose who had gastric contents in endotracheal aspirates) were young men, with a mean age of 27 years, whereas the majority of neurologic patients (includes two patients with metabolic coma, two patients with cardiac arrest, 14 patients with cerebrovascular ac-

TABLE I
Clinical Characteristics of Patients with Aspiration of Gastric Contents

	Overdose	Neurologic
Number	40	20
% Male	80.0	35.0
Mean age in years	27	68
% from community	77.6	22.4
Gastric pH: % of patients < 2.5	87.2	12.8
Initial a/A ratio	.533	.419
% Intubation	57.5	100.0
% Assisted ventilation	42.5	95.0
% Ultimate mortality	0	50

cidents, one patient with Guillain-Barre Syndrome, and one patient with encephalitis) were women, with a mean age of 68 years. Because of marked differences in these two subgroups, predisposing diagnosis was used as an independent variable in evaluation of the effects of corticosteroid treatment.

Based on x-ray findings and arterial blood gas studies, our patients could be divided into three subgroups of severity of illness: 1) Mild to moderate, 29 patients: 23 in the overdose group and six in the neurologic group. 2) Moderately severe, 19 patients: 11 in the overdose group and eight in the neurologic group. 3) Very severe, 12 patients: six in the overdose group and six in the neurologic group. There was no significant difference in severity of illness between the steroid and the control groups.

The mean time necessary for normalization of x-ray findings was significantly affected both by the predisposing condition and by corticosteroid treatment as shown in Figure 1. The mean normalization time for the treated overdose patients was 3.05 (1.57–4.53) days compared to 5.50 (4.02–6.98) days for the control group ($P < .05$), a reduction of 45 percent. The mean normalization time for the treated neurologic group was 5.67 (3.46–7.89) days compared to 8.42 (5.92–10.94) days in the control group ($P < .05$), a reduction of 33 percent. Parentheses indicate 95 percent confidence interval of the mean. A detailed analysis of x-ray findings in our patients is being prepared.

There was also a significant difference in the mean time for normalization of arterial blood gas studies between overdose patients in control and treatment groups (Fig. 2). Mean time for the treatment group was 3.20 (2.26–4.14) days compared to 4.79 (3.82–5.74) days for the control group ($P < .05$),

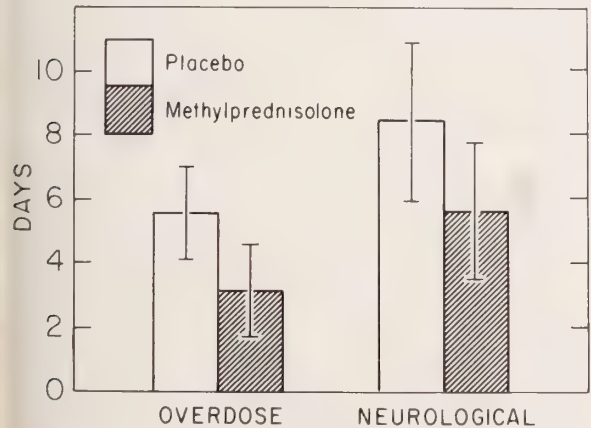


FIG. 1. Number of days for normalization of chest x-rays (mean with 95% confidence limits) after treatment with methylprednisolone sodium succinate 15 mg/kg body weight daily or placebo for three days.

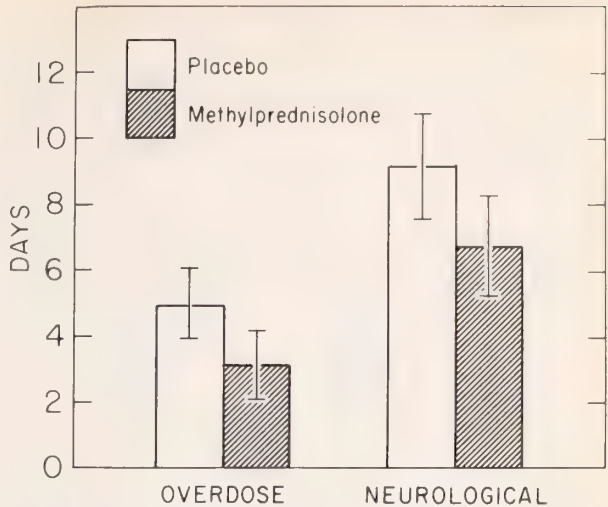


FIG. 2. Number of days required for normalization of arterial/Alveolar ratio.

a 33 percent reduction, whereas in the treated neurologic group it was 6.75 (5.27–8.23) days compared to 9.28 (7.70–10.9) days in the control group ($P < .05$), a 27 percent reduction.

In the overdose patient group, 28 of 40 required intubation and 22 required positive pressure ventilation. Thirteen patients in the treatment group and 15 patients in the control group required intubation ($P > .05$). Nine patients in the treatment group and 13 patients in the control group required mechanical ventilation ($P > .05$). Five patients received positive end expiratory pressure (PEEP), one in the overdose group and four in the neurologic group. In the latter group, two patients were in the treatment and control groups respectively. The duration of intubation and mechanical ventilation in the overdose group was significantly reduced in the treatment group as shown in Figure 3. The mean intubation time was 1.16 (.75–1.78) days for the treatment group compared to 2.60 (1.71–3.93) days for the control group ($P < .05$), a reduction of 55 percent. Ventilation time was .95 (.50–1.81) days for the treatment group and 1.88 (1.09–3.24) days for controls ($P < .05$), a reduction of 49 percent. A similar analysis could not be done for the neurologic group because of high patient mortality. Six of 11 patients in the treatment group and four of nine patients in the control group expired ($P > .05$).

A marked difference in length of stay in the intensive care unit in response to corticosteroid treatment between overdose and neurologic groups was found (Fig. 4). The mean length of stay of overdose patients was 3.25 (2.56–4.11) days in the treatment group compared to 4.43 (3.71–5.28) days in the control group ($P < .05$), a reduction of 26

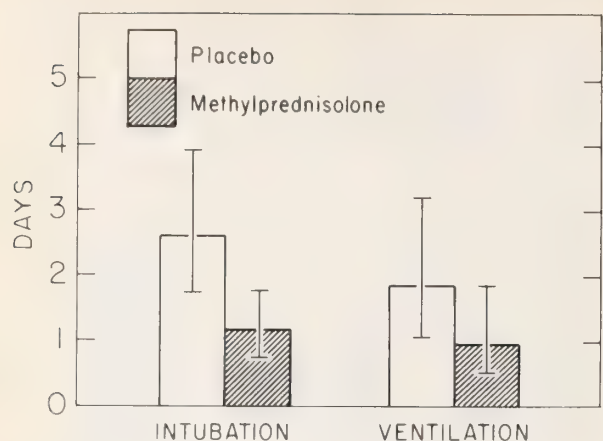


FIG. 3. Number of days required for intubation and ventilation for the overdose group.

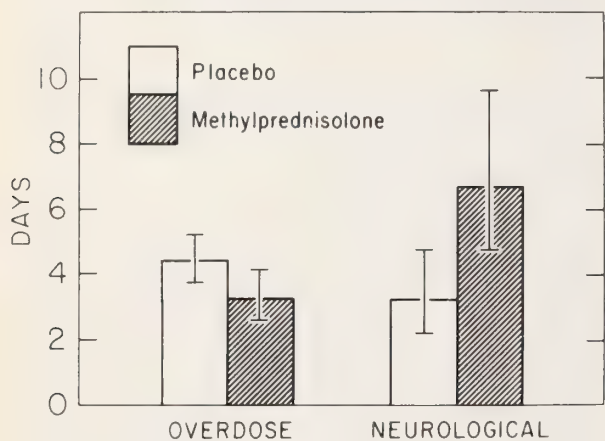


FIG. 4. Number of days in the intensive care unit.

percent. On the other hand the length of stay in the neurologic group was 6.77 (4.76–9.61) days on treatment compared to 3.22 (2.18–4.78) days in the control group ($P < .05$), an increase of 110 percent.

Table II summarizes complications seen in this study. There was no significant association between incidence of bacterial pneumonia, pleural effusion, hypokalemia, or sepsis and predisposing diagnosis with corticosteroid treatment ($P_{\alpha} > .05$, $P_{\beta} < .1$ for an expected double incidence of complications in the steroid group).

There was a significant difference in the proportion of patients having major gastrointestinal bleeding, between the overdose (10.5 percent) and neurologic groups (40.5 percent) ($P < .05$). However, there was no difference associated with corticosteroid treatment.

All ten deaths occurred in the neurologic group. Only three patients died during the three days of the study period. The cause of death in all patients was the underlying disease; one patient also had septicemia. There was no association between mortality and corticosteroid treatment.

TABLE II
Incidence of Complications*

Complications	Treatment (31)	Control (29)
G.I. Bleeding	13	13
major	7	6
minor	6	7
Hypokalemia	4	5
Sepsis	1	2
Pleural Effusion	2	6
Bacterial Pneumonia	3	5
Pneumothorax and Pneumomediastinum	1	0

* There was no significant association between any complication and corticosteroid treatment ($P > .05$).

Discussion

Following Mendelson's (1) initial description of aspiration of gastric contents, Hausmann and Lunt (2) first reported efficacy of corticosteroids in this syndrome. Subsequently, many reports appeared in the literature emphasizing a beneficial effect (8–12, 20–22). In recent experimental studies (4, 14–16) and clinical observations (13, 17) which are retrospective, have poor controls, and contain a limited number of cases, corticosteroids were not found to be of value. Although controversy exists, corticosteroids have been used in many centers for this syndrome. The rationale for steroid use rests on their antiinflammatory effects and ability to stabilize lysosomal membranes (23, 24), protect type II alveolar pneumocytes, prevent agglutination of platelets and leukocytes (25) and improve the peripheral release of oxygen from erythrocytes by shifting the oxyhemoglobin dissociation curve (26).

Supportive measures have been the mainstay in treatment of aspiration of gastric contents. Vigorous endotracheal suctioning and bronchoscopy are useful in removing particulate matter (27). Mechanical ventilatory support with or without positive end expiratory pressure (PEEP) may improve survival rate after aspiration (15, 16, 28). In addition to supportive function, Cameron and associates (28) observed other therapeutic benefits such as absence of pulmonary arterial vasospasm and thrombus formation in animals treated with assisted ventilation.

Our study concerns a restricted patient population, that is, drug overdose and neurologic disorders, with two thirds in the former category. In the overdose group, which had a large proportion of patients with a gastric aspirate pH less than 2.5 but with mild to moderate illness, the benefit of corticosteroids was statistically significant. Corticosteroid treatment resulted in a reduction of x-ray normalization time of 45 percent, arterial blood gas normalization time 33 percent, intubation time 55

percent, ventilation time 49 percent, and length of stay in the intensive care unit 26 percent. Wolf et al (17) reported a 56 percent reduction of ventilation days in patients who received corticosteroids.

The efficacy of corticosteroid treatment in the neurologic group with more severe illness is less clear. Steroids did not change the high mortality rate. Although there was a reduction of time for x-ray clearing (33 percent) and normalization of blood gas studies (27 percent), survivors remained in the intensive care unit 110 percent longer than did controls. This suggests that overall condition may deteriorate as a result of corticosteroid treatment. A controlled study of neurologic patients is needed to draw definite conclusions about this group.

We eliminated patients with heart failure and other underlying lung diseases such as bronchiectasis and diffuse interstitial lung disease in order to evaluate the role of corticosteroid treatment in patients with otherwise normal lungs. Many of our elderly neurologic patients had intercurrent illnesses which could have affected recovery.

Many patients with aspiration of gastric contents develop bacterial infection because of oropharyngeal contamination. Although the value of prophylactic antibiotics has not been established, many authors recommend their use (27, 29, 30). In this study, antibiotics were employed as part of a standardized treatment regime. Penicillin was given for community acquired disease since these patients usually have penicillin sensitive organisms (31–33). Clindamycin and gentamicin were given for hospital acquired aspiration since colonization is frequently due to penicillin resistant organisms (31–33). Only eight patients (13.3 percent) developed superimposed bacterial pneumonia, three in the treatment group and five in the control group. The difference was not statistically significant. Bynum and Pierce (13) reported a 26 percent incidence of pneumonia in their patients.

The occurrence of major gastrointestinal bleeding was not different in the treatment and control groups. The incidence of such bleeding in our respiratory care unit is 18 percent (unpublished data), a frequency similar to that reported in the literature (34). Major gastrointestinal bleeding, however, was more common in the neurologic group (40.5 percent) compared to the overdose group (10.5 percent). Corticosteroids did not increase the incidence of gastrointestinal bleeding.

Unilateral pleural effusion was noted in eight of our patients (13.3 percent), two in the treatment group and six in the control group. The difference was not statistically significant. We believe that chemical pneumonitis contributed to its develop-

ment. Pneumothorax and pneumomediastinum were seen in only one patient. This contrasts with a reported incidence of 38 percent (35).

The mortality rate in our study was 16.6 percent. All deaths occurred in the neurologic group. Reported mortality varies from 30–71 percent in clinical studies (4, 6, 9, 36, 37) and up to 100 percent in experimental studies (6, 22). All our patients died because of their underlying disease which predisposed to aspiration. Corticosteroid treatment did not affect mortality.

Conclusions

We conclude that corticosteroids in the form of methylprednisolone sodium succinate (Solumedrol®), 15 mg/kg body weight in three divided doses daily, for three days in young patients with drug overdose and mild to moderate aspiration pneumonia, hasten recovery without detrimental effects. They may have the same beneficial effects on recovery in severely ill elderly neurologic patients with other complicating medical illnesses, but may also contribute to morbidity by as yet poorly defined mechanisms. Corticosteroids do not improve the incidence of mortality in either patient group. The results may not be universally applicable to other groups of patients with aspiration of gastric contents. Thus, their use must be individualized and selective.

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Erratum

In Vol. 47, Issue 3, p. 289 in "Verruca Planavulgaris" by Dr. John Kwittken: The eighth criterion in Table II should read:

Enlarged cells with vacuolated cytoplasm and central shrunken or normal-sized nuclei in granular and upper malpighian layers.

Influence of Corticosteroid Treatment on Pulmonary Function After Recovery from Aspiration of Gastric Contents

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Abstract

Twenty-one survivors in a prospective investigation evaluating corticosteroids were studied after an acute episode of pulmonary aspiration of gastric contents. The patients were relatively young and healthy before the acute episode. Sixteen of the 21 patients were cigarette smokers. All except one aspirated after drug overdose. Most of the patients had mild to moderate pneumonia. Only one patient required positive end expiratory pressure. The mean follow-up time was 12 months. Nineteen patients were asymptomatic and 20 patients had normal chest x-rays at time of follow-up. The most frequent abnormalities of pulmonary function were significant reduction in maximal expiratory flow rates at 50 percent and/or 75 percent of expired vital capacity found in 17 patients. This obstructive abnormality was partially reversible in seven patients following inhalation of bronchodilators. The abnormalities in small airways of these patients are probably explained by both the high incidence of cigarette smoking and previous lung injury by gastric contents. There was no significant difference in long-term pulmonary function tests between those who received steroids or placebos during the acute episode of aspiration in this selected patient population.

The long-term sequelae following aspiration of gastric contents into human lungs are not well documented. However, a few recent studies have shown little residual impairment after recovery (1-4). The usefulness of corticosteroid treatment is controversial (5, 6). Recently in a double-blind controlled clinical trial we have shown that methylprednisolone hastened recovery without detrimental effects in young overdose patients with mild to moderate aspiration pneumonia during the acute phase of aspiration of gastric contents (7). There are no previous data assessing the role of steroids in modifying long-term sequelae of patients with aspiration. We report a long-term follow-up study comparing pulmonary function in patients who received corticosteroid treatment with those who received placebos.

Materials and Methods

During the period July 1976 to June 1978, 95 patients with aspiration of gastric contents were treated at the Mount Sinai Services-City Hospital Center at Elmhurst, New York. The first 35 patients were placed in a pilot prospective alternate corticosteroid study and the last 60 patients were placed in a double-blind controlled, clinical trial.

Of the 95 patients, 82 survived. All survivors were contacted by telephone; 21 patients (six from the pilot study) returned for follow-up studies. Twelve patients had received steroids.

The diagnosis of aspiration of gastric contents was based upon two sets of criteria. In 16 patients (includes three patients with narcotic overdose) diagnosis was made by observation of gastric contents during intubation or removal of gastric contents from the tracheobronchial tree. In the remaining five patients diagnosis was made on clinical grounds such as chest x-ray and arterial blood abnormalities after observed vomiting.

All patients received similar standardized treatment as follows: oxygenation, suctioning, careful administration of intravenous fluids, antacids, pen-

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icillin for community acquired aspiration or clindamycin and gentamicin for hospital acquired disease. Eighteen patients required mechanical ventilation and one patient required positive end expiratory pressure (PEEP). Patients treated within 24 hours after diagnosis of pulmonary aspiration received either methylprednisolone sodium succinate (Solumedrol®) 15 mg/kg body weight daily in three divided doses intravenously for three days or a placebo.

Table I lists clinical data, diagnosis, treatment, and complications. Each patient was evaluated clinically and his chest x-ray reviewed by the authors. Except for one individual (patient 11) who had past history of asthma in childhood, all patients were healthy without pulmonary complaints before the episode of aspiration.

The arterial/Alveolar ratios were calculated on the initial and daily arterial blood gases until normalization (a/A ratio > 0.8) by using the simplified alveolar air equation:

$$P_{AO_2} = P_{IO_2} - PaCO_2 \times \frac{1}{R}$$

with R , the respiratory exchange ratio, assumed to equal 0.8 (8).

Pulmonary function studies were done in the sitting position with standard techniques. Routine spirometry was performed with a dry spirometer and lung volumes were corrected to body temperature and pressure, saturated with water (BTPS). Airway resistance and functional residual capacity (FRC) were measured using the body plethysmograph method of DuBois and associates (9, 10). Maximum expiratory flow volume curves were recorded on an X-Y recorder (CPI Model 750). Carbon monoxide diffusing capacity (Dsb) was measured using the single breath method (11). Closing volume and nitrogen slope were determined from a modified single breath nitrogen test. Arterial blood gases were analyzed for pH, carbon dioxide tension ($PaCO_2$), and oxygen tension (PaO_2) using separate electrodes in a Radiometer blood gas analyzer. Predicted normal values were obtained from Boren and associates (12) and Goldman and Becklake (13) for lung volumes, Morris and associates (14, 15) for spirometry, Cherniack and Raber (16) for the flow

TABLE I
Clinical Data of Patients in the Steroid (1-12) and Control (13-21) Groups

Patient	Age/Sex	Height (inches)	Smoking (pack-years)	Overdose of	Gastric pH	Initial a/A Ratio	No. of Days till a/A > 0.8	Complications
1	41/F	63.75	60	Ethchlorvynol	3	0.47	10	Collapsed LLL
2	30/F	61.25	0	Amitriptyline, chlordiazepoxide	—	0.4	3	—
3	16/M	65.25	4	Alcohol	2	0.67	2	—
4	23/M	69	5	Phencyclidine	2	0.53	2	—
5	24/M	70.25	18	Diazepam, methadone	2	0.43	7	—
6	23/M	68	4	Secobarbital	2	0.3	3	—
7	25/M	71.25	8	Secobarbital and amobarbital	1	0.68	2	—
8	37/M	66	15	Amitriptyline	2	0.68	2	—
9	70/M	66	20	Diazepam, secobarbital, meprobamate	4	0.65	3	GI bleeding
10	36/F	63.5	22	Diazepam, alcohol	2	0.57	2	—
11	31/M	68.75	6	Heroin	3	0.30	8	Pneumothorax
12	25/M	69.75	12	Ethchlorvynol, meprobamate	1	0.55	4	—
Mean $\pm$ SEM	31.8 $\pm$ 4.0	66.89 $\pm$ 0.89			2.2 $\pm$ 0.3	0.52 $\pm$ 0.04	4.0 $\pm$ 0.8	
13	40/M	63.75	0	Unknown drug	2	0.22	5	Pleural effusion
14	38/F	62	24	None (alcohol withdrawal)	2	0.39	11	GI bleeding
15	37/F	62.5	25	Amitriptyline, diazepam	4	0.7	7	Pneumonia
16	67/F	59.75	0	Chlorpromazine, amitriptyline	2	0.5	6	—
17	37/F	59	0	Diazepam, secobarbital and amobarbital	1	0.5	4	Pneumonia
18	62/M	67.75	0	Phenobarbital, phenytoin	2	0.25	17	GI bleeding
19	44/F	66	60	Secobarbital	—	0.48	3	—
20	26/M	71	4	Haloperidol	2	0.56	17	Lung abscess
21	21/M	65	10	Heroin	2	0.11	8	—
Mean $\pm$ SEM	41.3 $\pm$ 4.7	64.08 $\pm$ 1.28			2.1 $\pm$ 0.3	0.41 $\pm$ 0.05	8.7 $\pm$ 1.8	

volume curves, Buist and Ross (17, 18) for the closing capacity and nitrogen slope, Ayers (19) for the single breath Dsb, Mellemaard (20) for the alveolar-arterial oxygen pressure difference ($P(A-a)O_2$).

The results of pulmonary function studies in control and treatment groups were compared by Student's T Test and Fischer's Exact Test. An alpha error (P_α) of less than 0.05 was considered significant. In addition, beta error (P_β) was computed assuming a difference between the means of the two groups in comparison to be greater than 20 percent. A P_β of less than 0.1 was considered significant.

Results

The mean age of patients in this study was 35.8 years. There were 13 men and eight women. Twelve patients received steroids and nine received placebos. The mean age of the treatment group was 31.8 years compared to 41.3 years for the control group ($P_\alpha > .1$). Eleven of 12 patients in the treatment group were smokers compared to five of nine in the control group ($P_\alpha > .05$).

The precipitating cause of aspiration was drug overdose except in one patient who had an alcohol withdrawal syndrome. The mean gastric pH was 2.2 in the treatment group compared to 2.1 in the control group ($P_\alpha > .05$). The mean initial arterial/Alveolar ratio was .52 in the treatment group and .41 in the control group ($P_\alpha > .05$). There was a significant difference between the mean normalization time of arterial blood gas studies (arterial/Alveolar ratio > 0.8) between subjects in the two groups. The mean normalization time for the treatment group was 4.0 ± 0.8 days compared to 8.7 ± 1.8 days for the control group ($P_\alpha < .05$). The mean time between hospitalization and follow-up study was 10.8 ± 2.7 months in the treatment group and 13.7 ± 2.7 months for the control group ($P_\alpha > .05$).

Nineteen patients were completely asymptomatic. Patients 10 and 19 were heavy smokers and had productive coughs for more than two years.

Chest x-rays were normal in 20 individuals and showed hyperinflation in one patient in the control group (patient 18).

The results of pulmonary function studies are summarized in Tables II and III and in Figures 1 and 2.

Lung Volumes, Spirometry, and Flow-Volume Curves

There was no statistically significant difference between the measurements of total lung capacity

(TLC), residual volume (RV), vital capacity (VC), forced expiratory volume in one second (FEV_1), and FEV_1/FVC ratio ($P_\alpha > .05$, $P_\beta < .1$) in treatment and control groups. There was also no significant difference between studies of the combined treatment and placebo groups compared with predicted normals ($P_\alpha > .05$, $P_\beta < .1$).

The instantaneous forced expiratory flow rate measured at 50 percent of vital capacity ($\dot{V}_{max50\%}$) was 84.4 ± 5.6 percent of predicted for the treatment group and 87.7 ± 11.7 percent of predicted for the control group. The instantaneous forced expiratory flow rate measured at 75 percent of expired vital capacity (that is, when 25 percent remained to be expired) ($\dot{V}_{max75\%}$) for the two groups respectively was 50.4 ± 6.2 percent and 61.6 ± 9.2 percent of predicted. The statistical difference between these two measurements is indeterminate for the size of this study ($P_\alpha > .05$, $P_\beta > .1$).

The mean $\dot{V}_{max50\%}$ and $\dot{V}_{max75\%}$ of the combined treatment and placebo groups were 85.8 ± 5.8 percent and 55.2 ± 5.3 percent, significantly decreased compared to predicted values ($P_\alpha < .05$). The $\dot{V}_{max50\%}$ and/or $\dot{V}_{max75\%}$ measurements increased significantly more than 20 percent in seven patients after bronchodilators (patients 8, 9, 11, 12, 18-20).

Airway Resistance

The airway resistance (R_{aw}) of every patient in this study was within normal limits.

Single Breath Nitrogen Test

The difference between the two groups for the ratio of closing capacity and total lung capacity (CC/TLC percent) and the nitrogen slopes ($\Delta N_2/L$) was statistically indeterminate ($P_\alpha > .05$, $P_\beta > .1$). However, there was no significant difference between the combined groups and the predicted normal ($P_\alpha > .05$, $P_\beta < .1$).

Gas Exchange

There was no statistically significant difference in the PaO_2 and the $P(A-a)O_2$ between the treatment group and control group ($P_\alpha > .05$, $P_\beta < .1$). The $P(A-a)O_2$ of the combined group when compared to the predicted normal was statistically indeterminate ($P_\alpha > .05$, $P_\beta > .1$). There was no statistical difference in the single breath carbon monoxide diffusing capacity (Dsb) between the treatment and control groups and between the combined groups and the predicted normal ($P_\alpha > .05$, $P_\beta < .1$).

TABLE II

Lung Volumes, Spirometry and Maximal Expiratory Flow Rates in the Steroid (1-12) and Control (13-21) Groups

Patient	Interval After Episode (Months)	TLC (% Pred)	RV (% pred)	VC (% pred)	FEV ₁ (% pred)	FEV ₁ / FVC %	$\dot{V}_{max50\%}$ (% pred)	$\dot{V}_{max75\%}$ (% pred)
1	23	82	54	92	99	84	90	51
2	1	101	101	94	104	88	110	69
3	15	93	88	84	87	88	73	71
4	1	99	70	99	100	81	117	86
5	7	104	54	110	109	81	93	78
6	1	104	66	103	98	77	88	69
7	1	120	108	115	104	73	73	41
8	17	83	68	83	85	82	104	43
9	9	98	112	96	103	73	67	41
10	20	105	88	107	101	75	66	35
11	27	107	177	79	70	69	54	44
12	8	114	76	115	110	76	78	61
Mean $\pm$	10.8	100.8	88.5	98.1	97.5	78.9	84.4	50.4
SEM	2.7	3.2	9.8	3.6	3.3	1.7	5.6	6.2
13	23	97	94	93	100	88	127	107
14	3	101	125	86	95	87	96	64
15	1	111	129	100	110	89	95	92
16	18	111	52	149	168	80	157	88
17	18	92	100	82	82	79	58	34
18	17	106	128	98	93	67	53	33
19	6	92	101	86	86	77	71	43
20	19	99	92	94	89	74	62	41
21	18	90	79	85	83	83	70	52
Mean $\pm$	13.7	99.9	100.0	102.0	100.7	80.4	87.7	61.6
SEM	2.7	2.7	8.4	11.7	8.9	2.4	11.7	9.2

FEV₁ forced expiratory volume in one second

FVC forced vital capacity

RV residual volume

TLC total lung capacity

 $\dot{V}_{max50\%}$ instantaneous forced expiratory flow rate at 50% of vital capacity $\dot{V}_{max75\%}$ instantaneous forced expiratory flow rate at 75% of vital capacity

Discussion

There have been few long-term studies of patients following aspiration of gastric contents. Sladen et al (21) reported one patient who had reduced lung volumes, decreased compliance, increased physiologic deadspace to tidal volume ratio, ventilation perfusion abnormality, and chest x-ray changes suggestive of interstitial fibrosis 163 days following aspiration. Recent reports, however, suggest that the majority of patients without preexisting lung disease are asymptomatic and physiologically either normal or nearly normal on follow-up examination. It has been suggested that corticosteroids may decrease the amount of fibrosis that follows the acute phase of the aspiration. Unfortunately, the role of corticosteroids in modifying the long-term outcome of pulmonary aspiration cannot be assessed because steroids were given to all patients in these reports (2-4).

We have studied 21 survivors after an acute episode of pulmonary aspiration of gastric contents in a prospective corticosteroid study. The mean age of patients was 35.8 years. All were healthy before this acute episode. The predisposing factor was drug overdose in all except one. Most of the patients had mild to moderate aspiration pneumonia. Only one patient required PEEP. The mean follow-up time was 12 months. Nineteen survivors were asymptomatic and 20 had normal chest x-rays at the time of follow-up. The most frequent pulmonary function abnormalities were significant reduction in maximal expiratory flow rates at 50 percent and/or 75 percent vital capacity found in 17 patients. This obstructive abnormality was partially reversible following inhalation of bronchodilators in seven patients.

In our study the differences between treatment and control groups were not statistically significant (that is, less than 20 percent) for lung volumes,

TABLE III

Airway Resistance, Single Breath Nitrogen Test, and Gas Exchange in Steroid (1-12) and Control (13-21) Groups

Patient	Raw cm H ₂ O/L/sec	CC/TLC % (% pred)	N ₂ Slope ΔN ₂ /L (% pred)	PaO ₂ mm Hg	P(A-a)O ₂ mm Hg	Dsb ml/mm Hg/min (% pred)
1	0.96	75	71	88	12	96
2	0.92	109	76	102	0	92
3	1.41	—	115	91	4	108
4	1.09	86	85	96	4	—
5	0.83	77	74	103	0	83
6	1.34	—	—	91	8	113
7	1.12	98	94	91	18	—
8	1.31	91	93	81	20	61
9	1.30	111	85	68	26	64
10	1.52	95	110	88	14	83
11	0.84	101	98	87	23	84
12	1.00	118	83	101	4	137
Mean ± SEM	1.14 0.07	96.1 4.5	89.5 4.3	90.9 2.8	11.1 2.6	92.1 6.6
13	1.57	93	90	87	21	150
14	1.42	—	—	95	10	79
15	0.72	—	80	91	14	73
16	1.76	63	90	109	6	109
17	1.59	106	95	105	2	84
18	1.39	105	90	74	25	62
19	1.62	108	91	80	15	75
20	1.10	102	82	90	8	98
21	1.09	—	163	88	9.5	86
Mean ± SEM	1.36 0.11	96.8 7.8	97.6 9.5	91.0 3.7	12.3 2.4	90.7 8.7

CC closing capacity
Dsb single breath carbon monoxide diffusing capacity
PaO₂ arterial oxygen tension
P(A-a)O₂ alveolar-arterial oxygen tension difference
Raw airway resistance

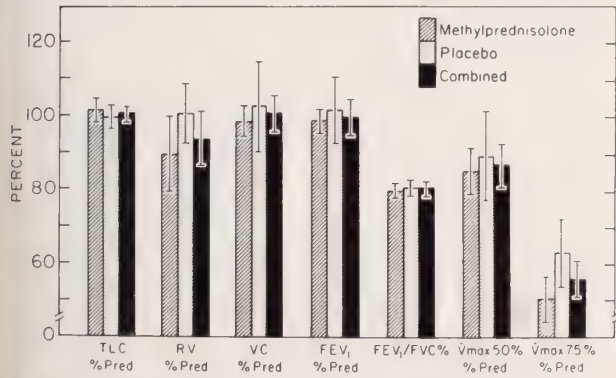


FIG. 1. Lung volumes, spirometry, and maximal expiratory flow rates at lower lung volumes in steroid, placebo, and combined group.

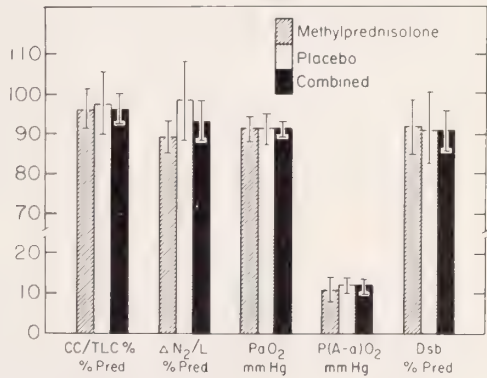


FIG. 2. Single breath nitrogen test and gas exchange in steroid, placebo, and combined group.

spirometry, P(A-a)O₂, and Dsb. However, for the V_{max}50%, V_{max}75%, and single breath nitrogen tests the differences between treatment and control groups are statistically indeterminate and could exceed 20 percent. Greater numbers of patients

would be required to answer this question. The mean lung volumes, spirometry, airway resistance, single breath nitrogen test, P(A-a)O₂, and Dsb of the combined group were within predicted normal range, and differences in these studies except P(A-

a)O₂ when compared to predicted normals were not statistically significant.

The reduction in $\dot{V}$ max at low lung volumes in the presence of normal FEV₁ suggests abnormalities in small airways. The small airway obstruction in our patients is probably explained by both the high incidence of cigarette smoking and previous lung injury by gastric contents. Peters and Ferris (22) demonstrated significant reduction in $\dot{V}$ max at low lung volumes in heavy smokers when compared with nonsmokers in a college-age group. Reversible airway obstruction is also known to develop after a variety of lung injuries including those that lead to the adult respiratory distress syndrome (23). The mechanism of this reversible airway obstruction is not known. Long-term follow-up of a larger population of nonsmokers may help separate the role of lung injury by gastric contents from the effect of cigarette smoking.

The usefulness of corticosteroid treatment during the acute stage in pulmonary aspiration of gastric contents is controversial. Recently in a double-blind controlled, clinical trial we showed that methylprednisolone sodium succinate 15 mg/kg body weight in three divided doses daily for three days, along with vigorous supportive treatment, hastened recovery without detrimental effects in young overdose patients with mild to moderate aspiration pneumonia. Corticosteroid treatment reduced chest x-ray and arterial blood gas normalization times but did not affect mortality or incidence of complications (7). In the present study we have shown that after recovery there was no clinical evidence of pulmonary fibrosis and no significant difference in long-term pulmonary function studies between those who received steroids or placebos during the acute episode in this selected patient population.

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Dermatitis

JOHN KWITTKEN, M.D.

Abstract

Microscopic interpretations of inflammatory skin diseases probably encompass the most difficult diagnostic problems encountered in dermatopathology. One reason is because the skin can react only in a limited number of ways, often the same way, to different stimuli. A rational approach to their interpretation based upon the primary site of the inflammatory response has been proposed. Eleven general categories have been outlined. Pathologic processes do not respect man-made boundaries and overlapping of categories occurs in many if not most diseases. Ambiguous terms have been defined and the abandonment of useless and improper ones has been suggested. Discussions of several controversial inflammatory dermatoses have been presented in an attempt to clarify some ambiguities and misconceptions regarding them. These include eczema, parapsoriasis, poikiloderma, pityriasis lichenoides and erythema multiforme.

Inflammation may be defined as a dynamic complex of cellular reactions that occurs in response to an injury or abnormal stimulation caused by a physical, chemical, or biologic agent. Perhaps the most difficult diagnostic problems encountered in dermatopathology involve the microscopic interpretation of inflammatory diseases or dermatitis. One reason is because the skin can react only in a limited number of ways, often the same way, to different stimuli. Too often, the histologic features are nonspecific and nondiagnostic. It is not the intent of this paper to detail the characteristic histologic features of inflammatory skin diseases. The purpose, rather, is to present a simple rational approach to the histologic interpretation of these diagnostic problems which will afford the clinician the maximum amount of information that can be gained from the biopsy. Also presented is an understanding of some ambiguous terms as well as suggested abandonment of seemingly useless and improper ones that are commonly used in microscopic descriptions of inflammatory skin diseases. Briefly discussed are viewpoints of several controversial inflammatory dermatoses. Many of the opinions set forth in this paper do not reflect prevailing thought. Nevertheless, correct or not, it is hoped that the thought and discussion generated herein will aid in

the understanding and solution of controversial issues as well as facilitate the diagnostic interpretation of any or many of the inflammatory dermatoses.

Classifications of dermatitis are readily available in well recognized textbooks of dermatopathology (1-4). Recently (5), classification based upon the pattern of the inflammatory reaction has been proposed. While the former classifications seem too broad and nonspecific, the latter appears too restrictive and cumbersome. It must be stated and understood from the onset that all of the characteristic histologic features of many cutaneous inflammatory diseases may not be present and often overlapping occurs with other diseases. In other words, pure packaging of disease by ingredients as occurs in a neighborhood grocery store often is not possible.

It is useful to place inflammatory skin diseases into eleven general categories (Tables I to XI), based upon the primary site of the inflammatory response. Included are some diseases which, although not basically inflammatory, have inflammatory components which often mimic histologically true primary inflammatory dermatoses. Conversely, excluded are some diseases which are basically inflammatory, but which present clinically and histologically as tumorous growths. *It cannot be over-emphasized that these tables represent nothing more than a general reference guide for the differential diagnosis of the inflammatory dermatoses.* Although most of the inflammatory skin diseases are listed, these Tables are neither com-

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TABLE I
Vasculitis

Primary	
Nonspecific	Lucio's phenomenon
Allergic granulomatosis	Necrotizing
Erythema elevatum diutinum	Polyarteritis nodosa
Erythema nodosum leprosum	Purpura pigmentosa chronica
Granuloma faciale	Temporal arteritis
Leukocytoclastic	Wegener's granulomatosis
Secondary	
Erythema induratum	Urticaria
Erythema multiforme	Viral exanthems
Erythema nodosum	Herpes simplex
Macular atrophy	Herpes zoster
Mucha-Habermann's disease	Varicella
Septicemia (bacterial)	Variola

TABLE II
Adnexitis

Hidradenitis	
Nonspecific	Fox-Fordyce disease
Miliaria	Hidradenitis suppurativa
Folliculitis	
Nonspecific	Folliculitis barbae
Acne conglobata	Folliculitis decalvans
Acne necrotica miliaris	Folliculitis keloidalis nuchae
Acne rosacea	Furuncle
Acne varioliformis	Impetigo Bockhart
Acne vulgaris	Perforating folliculitis
Alopecia areata	Perifolliculitis capitis abscedens et sufficiens
Alopecia cicatrisata	Ulerythema of eyebrows and cheeks
Alopecia mucinosa	
Disseminate infundibulo-folliculitis	

prehensive nor absolute. Pathologic processes do not respect man-made boundaries. Overlapping occurs, as previously stated, in many if not most of these diseases. Furthermore, not only are new inflammatory disorders being currently discovered, but also many fine dermatopathologists may have healthy disagreements with what this paper contends to be the primary site of the inflammatory response in the dermatoses listed.

The Table headings indicate the primary location of the inflammatory changes. Most of them are self-explanatory. The reader will note, except for vasculitis and adnexitis, the remaining Tables represent either individual levels or various combinations of levels of involvement. From the surface downward, these levels consist of epidermis, papillary cutis, reticular cutis and panniculus (subcutaneous tissue). In my opinion, sole epidermal inflammation is nonexistent. Diseases which involve primarily both the epidermis and papillary dermis are desig-

TABLE III A
Junction Dermatitis

Blistering	
Nonspecific	Miliaria
Acropustulosis of infancy	Nummular
Atopic	Papular urticaria
Bullosis diabetorum	Pemphigus
Bullous pemphigoid	Polymorphous light eruption
Candidiasis	Pompholyx
Cicatricial pemphigoid	Ritter's disease
Contact	Scabies
Darier's disease	Subcorneal pustular dermatosis
Dermatitis herpetiformis	Transient acantholytic dermatosis
Dominant ichthyosis congenita	Transient neonatal pustular melanosis
Electric burns	Urticaria pigmentosa
Epidermolysis bullosa	Viral diseases
Erythema multiforme	Hand-foot-and-mouth disease
Lyell's toxic epidermal necrolysis	Herpes simplex
Stevens-Johnson disease	Herpes zoster
Erythema toxicum neonatorum	Milker's nodules
Friction	Orf
Hailey-Hailey's disease	Varicella
Herpes gestationis	Variola
Hydroa aestivale	
Hydroa vacciniforme	
Impetigo	
Incontinentia pigmenti	

TABLE III B
Junction Dermatitis

Nonblistering	
Nonspecific	Parapsoriasis
Lichen planuslike	Pellagra
Psoriasiform	Pityriasis lichenoides
Atopic	Pityriasis rosea
Bloom's syndrome	Pityriasis rubra pilaris
Contact	Poikiloderma atrophicum vasculare
Dermatomyositis, erythematous edematous lesion	Poikiloderma congenitale
Dyskeratosis congenita	Porokeratosis
Erythema dyschromicum perstans	Prurigo nodularis
Erythema gyratum repens	Psoriasis
Erythema multiforme	Acrodermatitis continua
Exfoliative	Generalized pustular
Fixed drug eruption	Impetigo herpetiformis
Gianotti-Crosti syndrome	Localized pustular
Hartnup disease	Reiter's disease
Hyperkeratosis lenticularis perstans	Purpura pigmentosa chronica
Keratosis palmaris et plantaris	Recessive ichthyosis congenita
Lichen nitidus	Seborrheic
Lichen planus	Stasis
Lichen simplex chronicus	Sunburn
Lichen striatus	Superficial fungal infections
Lupus erythematosus, erythematous edematous lesion	Xeroderma pigmentosum
Necrolytic migratory erythema	

TABLE IV	
<i>Papillary Dermatitis</i>	
Nonspecific	
Dermal contact sensitivity reaction	
Drug reaction	
Erythema multiforme	
Photosensitivity reaction	
Toxic erythema	
Urticaria	

TABLE V	
<i>Reticular Dermatitis</i>	
Nonspecific	
Erythema annulare centrifugum	
Granuloma annulare	

TABLE VI	
<i>Papilloreticular Dermatitis</i>	
Nonspecific	
Acne rosacea	
Dermal contact sensitivity reaction	
Drug reaction	
Erysipelas	
Erythema multiforme	
Granuloma annulare	
Macular atrophy	
Pretibial pigmented patches in diabetes	
Sarcoidosis	
Urticaria	

TABLE VII	
<i>Junction-reticular Dermatitis</i>	
Nonspecific	Leishmaniasis
Angioimmunoblastic lymphadenopathy	Leprosy
Bromoderma	Lichen sclerosus et atrophicus
Cat-scratch disease	Lupus erythematosus
Chancroid	Mycobacterial infections
Coma induced bullae and sweat gland necrosis	Necrobiosis lipoidica
Deep fungal infections	Onchocerciasis
Dermatomyositis	Perforating granuloma annulare
Ecthyma	Polymorphous light eruption
Elastosis perforans serpiginosa	Radiation dermatitis
Erythema elevatum diutinum	Reactive perforating collagenosis
Erythema multiforme	Rhinoscleroma
Granuloma inguinale	Scabies
Interdigital pilonidal sinus	Septicemia (bacterial)
Iododerma	Sweet's syndrome
Kyrle's disease	Tattoo granuloma
	Treponemal infections

nated junction dermatitis. Here one finds, by far, the largest number of diseases. In a pandermatitis, all levels of the skin, including the panniculus, are involved.

Since the histology of mucous membrane is different from skin, use the terms superficial and deep when the upper and lower halves of the lamina

propria show inflammatory changes, respectively. The term junction is used when both epithelium and superficial lamina propria are primarily involved. Instead of the word dermatitis, utilize the name of the inflamed mucous membrane such as cheilitis, stomatitis, glossitis, gingivitis, conjunctivitis or vaginitis.

To indicate to the clinician the duration and degree of activity of the inflammatory process in *the tissue*, the terms acute, subacute, chronic and chronic active are used. An acute process is of relatively short duration, is associated with abundant edema and inflammatory cells, neutrophils often representing a major component, and ulceration and/or necrosis are often present. A chronic process is of relatively long duration, shows no edema, has a mixture of inflammatory cells with virtually no neutrophils, is usually associated with either a hyperplastic or atrophic epithelium or epi-

TABLE VIII	
<i>Panniculitis</i>	
Nonspecific	
Cold	
Erythema induratum	
Erythema nodosum	
Lupus erythematosus	
Rheumatoid nodule	
Sclerema neonatorum	
Scleroderma, early	
Subcutaneous dirofilariasis	
Subcutaneous fat necrosis of the newborn	
Subcutaneous granuloma annulare	
Subcutaneous nodular fat necrosis in pancreatitis	
Subcutaneous sarcoidosis	
Superficial migratory thrombophlebitis	
Weber-Christian disease	

TABLE IX	
<i>Panniculoreticular Dermatitis</i>	
Nonspecific	
Scleroderma	
Erythema induratum	

TABLE X	
<i>Panniculo-papillo-reticular Dermatitis</i>	
Nonspecific	
Autoerythrocyte sensitization	
Cryoglobulinemia	
Erysipelas	
Erythema nodosum	
Foreign body reactions	
Granuloma annulare	
Granuloma faciale	
Sarcoidosis	
Scleroderma, late	
Urticaria (angioedema)	

TABLE XI
Pandermatitis

Nonspecific
Acrodermatitis chronica atrophicans
Actinic reticuloid
Actinomycosis
Anthrax
Arthropod bite
Buruli ulceration
Chondrodermatitis nodularis helioides
Crohn's disease
Dermatomyositis
Lupus erythematosus
Maduromycosis
Mycetoma
Necrobiosis lipoidica
Nocardiosis
Perforating granuloma annulare
Pyoderma gangrenosum
Scleroderma
Tularemia

dermis and may or may not show varying degrees of fibrosis. A subacute process lies intermediate between an acute and chronic process in both duration and degree of activity. A chronic active process shows features of both a chronic and a subacute or acute process. *It is important to note that these terms do not necessarily reflect the duration or degree of activity of the clinical course.* For example, a biopsy of an involuting lesion in a patient with a recurrent flare-up of Mucha-Habermann's disease may show chronic changes. Remember that pathology is dynamic. A biopsy represents a moment in time of an ever-changing inflammatory process.

The terms focal or spotty and diffuse designate the pattern of the inflammatory changes in a particular category. Focal and spotty indicate small area involvement and diffuse refers to involvement of virtually the entire area in a particular category. The terms patch, patchy, nodule and nodular should not be used in microscopic descriptions. They should be utilized only for clinical descriptions of primary skin lesions.

Virtually all skin biopsies, whether of normal or inflamed tissue, show perivascular inflammatory cell accumulations of varying amounts. Their presence does not necessarily indicate inflammation. For this reason, it is meaningless and incorrect to use the term perivascularitis and perivascular dermatitis. By the same token, a so-called perifolliculitis is nonexistent and it is recommended here that this term be abandoned. When perifollicular inflammatory cell infiltrates are seen, they are always associated with a folliculitis.

The term granuloma is often used clinically for a purely histologic connotation of a pathologic pro-

cess which may present itself in the form of a variety of primary skin lesions (macule, patch, papule, plaque and nodule). This term designates a chronic inflammatory process of varied etiology (biologic, chemical or physical) characterized by the presence and preponderance of mononuclear cells (epithelioid cells, histiocytes, macrophages and lymphocytes) derived from the primitive reticular cell, with or without the presence of multinucleated giant cells, often, but not necessarily so, arranged in discrete, compact aggregates. Although the term granulomatous inflammation has basically the same meaning, in this report its use is reserved for a loosely arranged inflammatory cell infiltrate always associated with the presence of multinucleated giant cells and in which neutrophils and/or eosinophils may represent a prominent component.

Acantholysis denotes the process by which epidermal or epithelial cells become separated from each other in the lumen of a blister located above the basement membrane zone. Under light microscopy, the pathogenesis cannot be appreciated and may be variable. The free floating cells, occurring singly or in clusters, are designated acantholytic cells. Acantholytic cells occur in a variety of conditions which include pemphigus vulgaris, pemphigus vegetans, pemphigus foliaceus with its clinical variant pemphigus erythematosus, a wide range of dyskeratotic and acantholytic diseases occurring as incidental findings in various dermatoses, viral diseases and many nonspecific inflammatory and blistering dermatoses.

In microscopic descriptions of cutaneous inflammation, a physician often comes across the term round cell. This is an incorrect, nonspecific term intended to designate a nonexistent cell type and should be totally deleted from medical terminology. It is apparently intended to indicate nuclear shape and not cell shape. A variety of cells, inflammatory and neoplastic, may have round nuclei. These include lymphocytes, histiocytes, primitive reticular cells, macrophages, epithelioid cells, monocytes, leukemic and lymphomatous infiltrates and benign and malignant (primary or metastatic) epithelial neoplasms. To distinguish these cells from those having more than one nucleus, identifiable or not, inflammatory or neoplastic, refer to them as mononuclear cells. Although technically incorrect, neutrophils or eosinophils are not included under this term even though they possess only one nucleus.

Lichen planuslike and psoriasiform dermatitis are useful microscopic terms describing histologic characteristics resembling lichen planus and the typical full-blown stage of psoriasis, respectively. Their usage should be restricted to these meanings only.

Lichenoid, an ambiguous and incorrect term, should be abandoned.

Restrict the term vasculitis to those diseases in which histologic inflammation of a blood vessel wall is associated with evidence of organic damage to its wall. The essential criteria for vasculitis are either pure or mixed inflammatory cell infiltrates which include neutrophils, eosinophils and mononuclear cells involving either partial or entire thickness of a blood vessel wall associated with one or more of the following changes: perivascular hemorrhage, leukocytoclasia, multinucleated giant cells, mural necrosis (most often fibrinoid), thrombosis and mural fibrosis or hyalinization. The presence of any of these changes in the absence of mural inflammatory cells does not constitute a vasculitis. Conversely, the presence alone of nongranulomatous inflammatory cell infiltrates within and around blood vessel walls with or without endothelial swelling does not warrant a diagnosis of vasculitis. As can be seen from Table I, vasculitis may be either the primary disease or an accompaniment of other diseases.

There are several inflammatory skin diseases having characteristic clinical features which are reflected by nonspecific histologic changes. These include eczema, parapsoriasis, poikiloderma, pityriasis lichenoides and erythema multiforme. Because of this nonspecific clinicopathologic correlation, confusion and doubt about their actual existence have developed. A brief discussion of these diseases will follow in the hope of clarifying some ambiguities and misconceptions regarding them.

The term eczema is used often for so many dermatoses that its true meaning is frequently vague or lost. In its purest and simplest sense, eczema is a junction dermatitis of varied etiology characterized early by erythema, papules, vesicles, oozing and crusting, often followed later by scaling, lichenification and pigmentation. This term should be restricted to clinical usage only, since the histologic features are entirely nonspecific ranging from acute to chronic inflammatory changes.

The term parapsoriasis should be restricted to two entities characterized by chronic, asymptomatic scaling macules, patches, papules and plaques. These are parapsoriasis en plaques and variegata. The latter form often develops into poikiloderma atrophicum vasculare which will be discussed more fully later. Histologically, one observes in parapsoriasis en plaques and early lesions of variegata, a subacute to chronic nonspecific junction dermatitis. While the majority of cases remain as benign processes, a significant number of the former and some late poikilodermatous lesions of the latter develop into mycosis fungoides. Hence, in

a general sense, these lesions represent premalignancies. Some cases, however, probably represent an incipient stage of mycosis fungoides, either ab initio or developing from parapsoriasis, which could not be appreciated under light microscopy. In other words, these represent true pseudobenignities. There is no so-called premycotic lesion. A lesion is either benign or malignant. To advocate the existence of an "in-between" stage is merely an attempt to hide our human diagnostic failings.

Poikiloderma is a general term designating a group of diseases which has in common the clinical picture of atrophy, telangiectasia, and retiform variegated hypopigmentation and hyperpigmentation which is manifested histologically by epidermal atrophy, vascular ectasia and melanin incontinence. Included in this group are poikiloderma congenitale, Bloom's syndrome, dyskeratosis congenita, Hartnup disease, poikiloderma of Civatte, chronic radiodermatitis, poikiloderma atrophicum vasculare, mycosis fungoides, systemic lupus erythematosus and dermatomyositis. The last three diseases enumerated may develop within poikiloderma atrophicum vasculare. The histologic features show additionally either a nonspecific chronic or subacute junction dermatitis and/or superimposed specific diagnostic changes such as intraepidermal nests of lymphoma cells in mycosis fungoides, radiation fibroblasts in radiodermatitis, and liquefaction degeneration with fibrinoid change of the collagen in systemic lupus erythematosus and dermatomyositis. If the histologic features are not diagnostic, it is preferable to diagnose the lesion as poikiloderma.

Chronic, asymptomatic scaling papules, formerly designated as guttate parapsoriasis, should be classified under the terminology of pityriasis lichenoides. Histologically, they represent a nonspecific junction dermatitis which may be subclassified into chronic, subacute and acute forms. The acute form represents Mucha-Habermann's disease which may on occasion show microscopic features of a vasculitis or pseudolymphoma (lymphomatoid papulosis).

Erythema multiforme is an acute, self-limited, often recurrent inflammatory dermatosis of varied etiology showing a multiplicity of both clinical and histologic features as the name well indicates. Clinically, one may see in the same patient wheals, erythematous macules, papules and blisters as well as so-called target or iris lesions. Obviously, the histologic features are variable and depend upon the stage and type of primary lesion biopsied. The earliest change is a papillary or papilloreticular dermatitis corresponding to the wheals and erythematous macules observed clinically. Progression to

a junction or junction-reticular dermatitis with or without blistering gives rise to the remaining clinical lesions enumerated. There is often an associated vasculitis. Stevens-Johnson disease and Lyell's toxic epidermal necrolysis represent severe variants. Ritter's disease is an entirely different entity and should not be considered a variant of erythema multiforme.

Summary

A rational approach and guide for the diagnostic interpretation of inflammatory skin disorders has been proposed, based upon the primary site of the inflammatory response. Definitions of some ambiguous terms have been presented and the abandonment of others has been suggested. Viewpoints on

several controversial inflammatory diseases have been discussed.

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ERRATUM

In the article "Rheumatic Disease in the Elderly," (page 178 March/April 1980) Geriatrics issue under the Treatment section: the sentence should read 2) Avoid systemic corticosteroids as being valueless in osteoarthritis **NOT** osteoporosis.

Effect of Parathyroidectomy on Exocrine Pancreatic Secretion

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Abstract

The effects of parathyroidectomy on the steady state, secretin-induced pancreatic exocrine secretion were studied in a group of dogs. Free flow of pancreatic juice was obtained by direct cannulation of the main pancreatic duct (the minor duct being ligated). A gastric fistula prevented the entry of gastric acid into the duodenum. Parathyroidectomy caused significant reduction in the secretin-induced pancreatic secretion of volume and bicarbonate concentration, reciprocal change in chloride, and minute change in total protein concentration. These results were not dependent on calcium blood level, and were not affected by calcium administration to the hypocalcemic parathyroidectomized dog.

It is suggested that these effects of parathyroidectomy may reflect the direct involvement of the parathyroid hormone in the control mechanism of pancreatic exocrine secretion.

Introduction

Gastrin, secretin and CCK-PZ are the three prime gut hormones that regulate pancreatic secretion, but accumulated evidence suggests that other gastrointestinal polypeptides are involved as well. Neural mechanisms play an important, although not so succinct, role in the over all control of exocrine secretions (1). Clinical and experimental evidence indicates that calcium participates in regulating pancreatic function (2). It has been noted that chronic hypercalcemia associated with hyperparathyroidism predispose to the development of pancreatitis (3).

Calcium homeostasis is regulated mainly by parathyroid hormone (PTH) and calcitonin (CT). Although these two hormones affect pancreatic secretion indirectly via their influence upon calcium and gastrin levels (4), little evidence exists as to the possibility that they directly exert effect on pancreatic secretion.

This study was undertaken in order to test the effect of parathyroidectomy on the exocrine pancreatic secretion.

Materials and Methods

Under pentobarbital anesthesia (30 mg/kg), a group of dogs (weighing 15 to 25 kg) were prepared with chronic duodenal fistulae. The duodenum was mobilized, the accessory pancreatic duct ligated and divided, and a modified metal Thomas Cannula (5) was inserted in the duodenum, opposite the main pancreatic papilla. A gastric cannula was placed in the most dependent part of the stomach. At least three weeks were allowed to elapse for recovery. Each animal was deprived of food but not water, for 18 hours prior to each observation. The duodenal and gastric cannulas remained open during each experiment. Intravenous Saline (0.9 N) was given at a constant and fixed rate during all experiments. The main pancreatic duct was cannulated with a glass cannula and the pancreatic juice collected in consecutive ten minutes specimens under oil.

Secretin (Boots) was administered by continuous intravenous infusion (0.03 lu/kg/min) until the volume rate of secretion was constant for at least 30 minutes; this represented the control phase.

Samples of pancreatic juice were collected for measurements of volume and concentration of bicarbonate, chloride and total protein. Chloride was determined by the technique described by Schales (6). Bicarbonate was determined by the PH/Blood Gas Analyser, Model 413. Total protein was measured by the Biuret technique.

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In addition, blood samples were taken to determine serum calcium level. After three such observations on each animal, the dogs underwent total parathyroidectomy (verified by histologic examination). The dogs were then examined daily for the appearance of hypocalcemic tetany, and daily blood samples for calcium were drawn. Parenteral calcium was added when necessary. After parathyroidectomy, the same series of observations, and pancreatic juice collection and sampling was repeated, each dog serving as its own control.

Results

Parathyroidectomy produced a significant reduction in volume secretion and bicarbonate concentration, with reciprocal change in chloride and insignificant change in total protein concentration and out put. Summarized results of volume and bicarbonate changes, for all dogs, before and after parathyroidectomy are presented in Figure 1 and 2, respectively.

The mean volume secretion after a steady state was reached in the control studies was 5 cc, and mean bicarbonate concentration was 120 mEq/lit. After parathyroidectomy, the mean steady state volume secretion decreased by 40% and the mean bicarbonate concentration decreased by 30%.

The mean preoperative calcium blood level was 9 mg%, and the mean post-parathyroidectomy calcium level was 4.5 mg%.

Parenteral administration of calcium gluconate to the hypocalcemic animal did not change the rate of flow nor the bicarbonate concentration of pancreatic secretion.

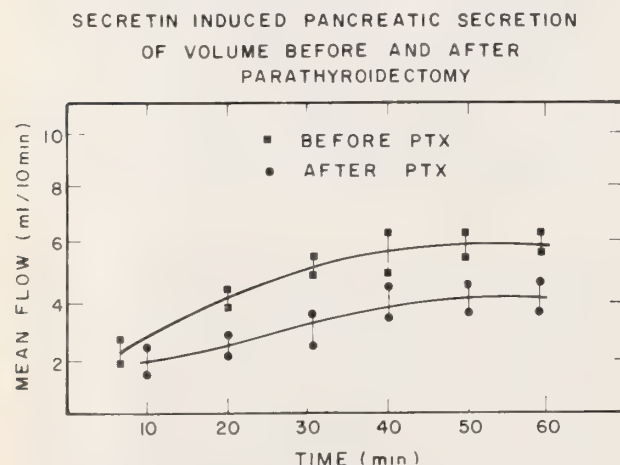


FIG. 1. Secretin-induced pancreatic secretion of volume and bicarbonate before and after parathyroidectomy. Each mark represents the mean results of three experiments performed on one dog. Secretin dose of 0.03 u/kg/min.

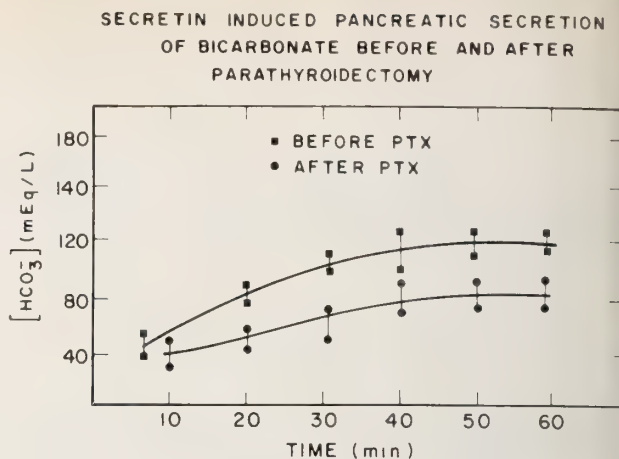


FIG. 2.

Discussion

Our results indicate that parathyroidectomy causes significant decrease in the secretin-induced pancreatic secretion of volume and bicarbonate concentration in dogs. The minute change in protein output and concentration can be attributed to the change in volume secretion. These results occurred in the absence of the gastric acid mediated duodenal secretin mechanism. There is a possibility that the post-parathyroidectomy changes in volume and bicarbonate are due to decreased PTH level in the plasma.

PTH is a polypeptide consisting of 84 amino acids. Its main mode of action at the subcellular level is to increase the amount of cyclic-AMP and change the direction of calcium flux (7, 8). The association of chronic hyperparathyroidism and pancreatic disease is well known (3, 9-11), but there is still insufficient explanation for this phenomenon (12).

There are two indirect effects of PTH on pancreatic exocrine secretions. The main effect is exerted by a decrease in calcium blood level (12), but our results demonstrate that the decrease in pancreatic secretion after parathyroidectomy is not dependent on calcium blood level, since calcium administration to the hypocalcemic dog did not alter the rate of secretion.

Another indirect effect of PTH, is through elevation of gastrin level in the blood as was shown by several authors (4, 13, 14), but the influence of gastrin on pancreatic volume secretion was less prominent than its effect on enzymes secretion (1, 14), and was shown to be dependent on gastric juice entering the duodenum (14), which was not the case in our experiments. It is possible that PTH, because

of its polypeptide structure, has a direct action on pancreatic secretion which might resemble the action of other gastrointestinal polypeptides (like secretin for example).

Another possibility is that PTH potentiates secretin activity. This hypothesis could be tested by studying the effect of PTH on basic (nonsecretin-stimulated) pancreatic secretion. Technically, this was impossible to study in our model, since we performed all our experiments on anesthetized dogs, in which state the basic pancreatic secretory values are very low (due to central nervous inhibition) and difficult to measure accurately (15, 16).

Although our results suggest a possible direct effect of PTH on pancreatic exocrine secretion, few problems still remain requiring further assessment, for example, the inclusion of antrectomy in the experimental model in order to completely exclude the possible role of gastrin. Another factor which would add more information would be the measurement of PTH blood levels during the experiments. These possibilities need further investigation.

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Mediastinal Histoplasmosis

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Abstract

Mediastinal histoplasmosis is now known to exist in two forms: an early or acute stage characterized by the presence of caseous or fibrocaseous lymph nodes within the mediastinum, and a late or chronic stage by fibrous mediastinitis. The term mediastinal granuloma was originally introduced to define the collection of caseous mediastinal nodes at a time when no specific organisms could be identified. Mediastinal granuloma was therefore considered an entity in itself of no known etiology. However, recent experience as well as improved staining techniques have shown histoplasmosis as the cause of mediastinal granuloma in approximately 85% of the cases and fibrous mediastinitis as the end stage of this disease spectrum. This paper will discuss the radiographic and clinical presentations of mediastinal granuloma and its evolution to fibrous mediastinitis. The findings vary according to the stage and depend upon which mediastinal structures are involved.

Materials and Methods

A total of 20 patients with varying stages of mediastinal histoplasmosis were reviewed at the University of Tennessee Center for Health Sciences. Fifteen patients manifested features consistent with the early form, and the remaining five demonstrated mediastinal fibrosis. The ages of the patients with early involvement ranged from 2 to 50 years with an overall average age of 13 years. The patients manifesting chronic disease were somewhat older, with ages ranging between 14 and 28 years with an average of 20 years.

Early Phase-Mediastinal Granuloma

H. capsulatum exists in soil, old chicken coops, pigeon, bird and bat guana. It is a highly communicable disease that is contracted following the inhalation of the fungal spores. After inhalation, the fungal spores are phagocytized by intra-alveolar macrophages and then multiply rapidly and germinate into yeast forms. The organism at this time appears as a round to oval budding yeast-like struc-

ture that varies from 2 mm to 4 mm in diameter. The fungus induces a primary pneumonia that soon transforms into a granulomatous reaction resembling tuberculosis. As in tuberculosis, drainage occurs into the regional hilar nodes and results in nodal enlargement and the formation of a primary complex. Nodal disease with enlargement persists for periods well after the primary pulmonary infection has healed. The lymph nodes histologically demonstrate marked hypertrophy of epithelioid cells and macrophages. Epithelioid or histiocytic granulomas are formed with central necrosis and giant cell development.

Most patients in this phase are asymptomatic. Cough and chest pain are, nevertheless, often encountered. Less common findings include fever, dyspnea, weight loss, dysphagia and hemoptysis.

Radiographic Features

Mediastinal Nodes

Radiographic evidence of enlarged mediastinal nodes is the most significant finding and was encountered in nine or 60% of the patients with mediastinal granuloma. The enlarged nodes occur most commonly on the right side of the mediastinum and an enlarged right paratracheal node is the most frequent finding (1). Five of the nine patients with lymphadenopathy demonstrated this manifestation alone. Predilection for the right side

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is apparently related to the higher incidence of granulomatous pneumonia involving the right lung as well as the preferential drainage of both lower lobe lymphatics to the right side.

The right paratracheal node is seen radiographically as a well defined oval or slightly nodular mass adjacent to the trachea (Fig. 1). Hilar adenopathy, seen as unilateral or bilateral hilar enlargement, is also not unusual (Fig. 2). Diffuse mediastinal involvement, subcarinal nodes, or combinations of all the above have also been encountered. Radiographs using high kilo-voltage technique or tomographic studies of the mediastinum better define the size and extent of the nodes. The disease is usually more extensive than is apparent on the routine radiograph (Fig. 2).

Esophagus

The esophagus is commonly indented and displaced by adjacent enlarged nodes. This feature is frequently clinically silent but may occasionally be associated with dysphagia. Although dysphagia is uncommon, it was the presenting symptom in two of our four patients who manifested esophageal involvement. The radiographic changes alone noted on the esophagram suggested the correct diagnosis. Dysphagia occurs most frequently when the sub-



FIG. 1. Paratracheal Lymphadenopathy. A large right sided oval shaped mass is located adjacent to the lower part of the trachea.



FIG. 2. Diffuse Adenopathy. Bilateral hilar nodal enlargement is noted. In addition to a right paratracheal lymph node, subcarinal nodes were also apparent on tomography.

carinal nodes are involved since the esophagus and the nodes are trapped between the heart, the stem bronchi, and the spine (Fig. 3A). The routine chest radiograph can be nonrevealing although careful scrutiny of the radiograph, high kilo-voltage technique, and/or tomography, will substantiate the presence of enlarged subcarinal nodes. The esophagus is moderately displaced and demonstrates a characteristic smooth extrinsic indentation with normal, intact mucosa (Fig. 3B). In most cases, the clinical and radiographic alterations regress without specific therapy. Unfortunately, one patient in our series was quite ill and underwent a thoracotomy with the possible diagnosis of a mediastinal abscess (Fig. 4).

Pericarditis

Pericarditis has not been considered a common complication of early mediastinal histoplasmosis. Approximately 31 cases have been recorded in the English literature (2). The incidence is probably much higher since we encountered three cases in one year. The onset of histoplasma pericarditis is somewhat insidious and consists of a prodromal period lasting for approximately four to five weeks duration with features of fever, rhinorrhea, cough, and malaise (2). The radiographic features at this time are obvious although nonspecific. The cardiac silhouette is either markedly enlarged or in the process of enlarging and demonstrates all the changes associated with pericardial effusion (Fig.

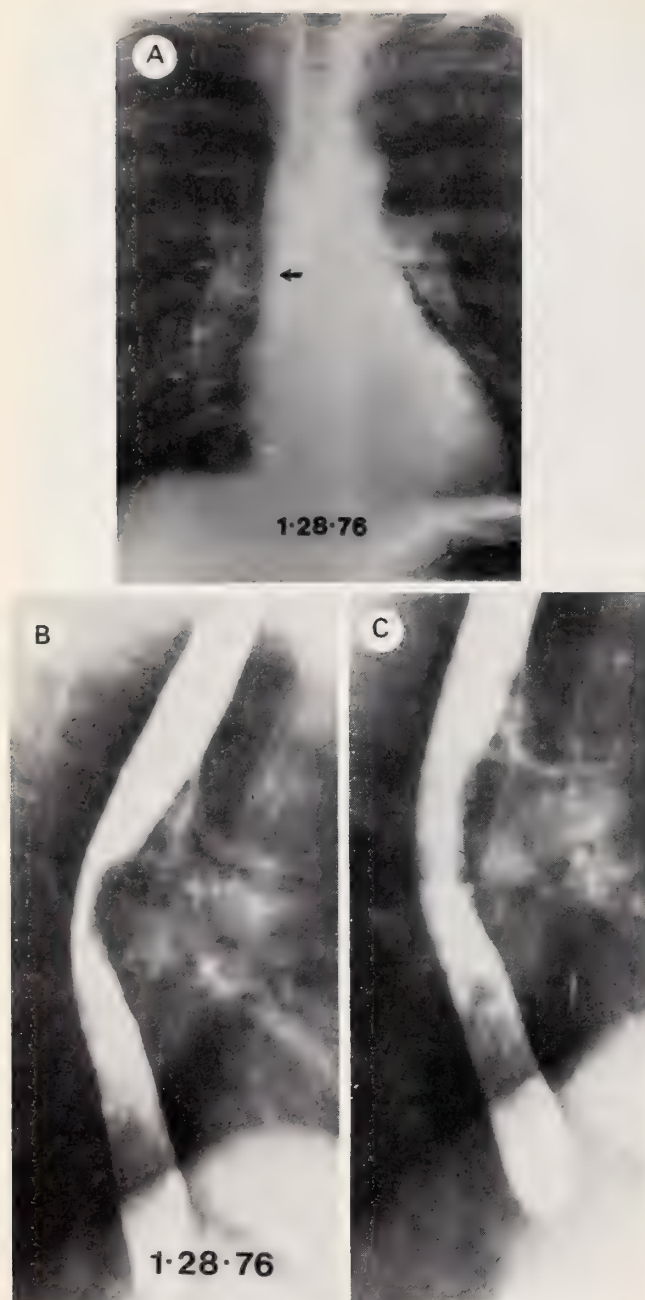


FIG. 3. Esophageal Involvement. A 31-year-old patient with dysphagia. 3A. Routine chest examination revealed a vague soft tissue mass within the mediastinum. (Arrow) 3B. The esophagus is displaced and smoothly indented anteriorly at the junction of its mid and distal thirds. 3C. One month later the esophageal changes are regressing.

4). The presence of associated lymphadenopathy and/or pneumonitis suggests a granulomatous process as the cause of the effusion. Enlarged nodes alone exclude a viral or idiopathic pericarditis; however, the possibility of an underlying lymphoma cannot be discounted.

The diagnosis of histoplasmosis as a cause of the pericarditis is established by inference since it is almost impossible to isolate the organism from the pericardial fluid. The recognition of certain clinical findings, for example, positive skin and/or serologic reaction may be sufficient. The prognosis is excellent despite the protracted period of weeks to months (average duration 2.5 months) required for convalescence (2). Constrictive pericardial disease is an infrequent complication, particularly when compared to the high incidence following tuberculous or coxsackie pericarditis. Early calcific deposition within the pericardium occurred in one patient approximately one year after the onset of the disease (Fig. 4D), although no clinical evidence of constrictive disease was apparent.

The pathogenesis of pericarditis associated with histoplasmosis is unclear. It may represent an extension of the disease from the adjacent lymph nodes. However, a hypersensitivity response similar to that of Dressler's Syndrome appears more plausible since there is a high incidence of associated pleural effusion, an occurrence that by itself is unusual in histoplasmosis.

Bronchial Compression

The mediastinal nodes usually remain anatomically distinct and rarely cause respiratory difficulty. However the trachea and bronchi are occasionally compressed and produce symptoms when they are sufficiently narrowed or when the nodes penetrate the bronchus producing endobronchial disease. The resulting symptoms and corresponding radiographic findings are caused by obstructive emphysema or by atelectasis (Fig. 5). Suspicion of histoplasmosis under these circumstances depends upon the recognition of juxtaposed enlarged lymph nodes.

Superior Vena Cava Syndrome

The superior vena cava is easily compressed. Its proximity to nodes located high and on the right of the mediastinum accounts for its frequent involvement. Pate noted this syndrome in four patients under ten years of age with mediastinal histoplasmosis (4). Although dilated and obstructive vessels are apparent, an effective collateral circulation develops quickly and few symptoms become clinically obvious.

Chronic Fibrosing Mediastinitis

The chronic or late stage of mediastinal histoplasmosis consists mainly of an accumulation of

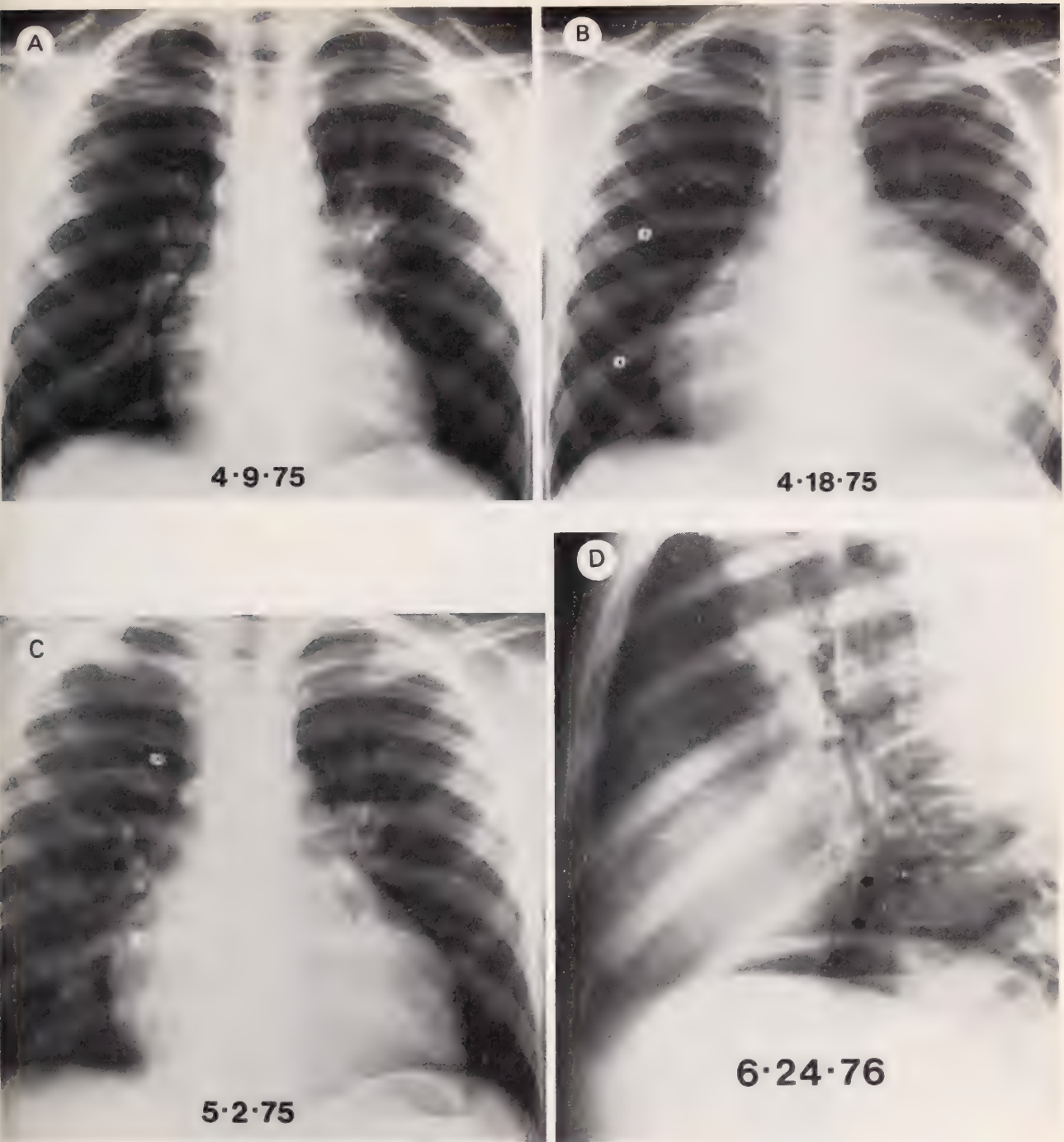


FIG. 4. Pericarditis. Fig. 4A. Initial chest examination (4/9/75) reveals left hilar nodal enlargement and a slightly prominent heart. Fig. 4B. Nine days later (4/18/75), the heart is markedly enlarged and demonstrates the classic water bottle configuration of pericardial effusion. Fig. 4C. Subsequent examination three weeks later (5/2/75) shows marked decrease in the amount of pericardial fluid. Fig. 4D. One year later (6/24/75). Early calcific deposition within the pericardium is occurring.

fibrous tissue which initially remains in proximity to the involved nodes but eventually may involve all contiguous mediastinal structures. The collagen develops adjacent to the diseased nodes and as a result affects the right side of the mediastinum

initially as in the early form of the disease. The developing fibrosis is quite dense and the underlying granulomatous component may not be conspicuous on the radiograph without the presence of obvious nodal calcification. Moreover, the causative



FIG. 5. Endobronchial Disease. Atelectasis of the right middle and lower lobes due to endobronchial disease within the bronchus intermedius is present. Large irregular nodes are also apparent on the left.

organisms are deeply buried within the fibrotic mass and are therefore almost impossible to identify. The major clinical and radiographic findings are variable and reflect the fibrotic process and its effect upon the neighboring mediastinal structures.

Radiographic Features

Chest Radiograph

The routine chest film in some cases shows relatively few findings. Varying degrees of mediastinal widening with an irregular ill-defined margin is nevertheless the principle manifestation of fibrous mediastinitis (Figs. 6A, 7A). This is often localized to the right side but can involve the entire mediastinum. When the mediastinal vessels become obstructed, the lungs show increased interstitial changes due to distended veins and lymphatics (Fig. 7A). Interstitial fibrosis eventually forms, adding to the parenchymal opacities. The latter is the result of chronic congestion. The interstitial changes are also commonly localized to the right lung, although bilateral changes eventually supervene.

Tracheobronchial Tree

Circumferential stenosis of the trachea and bronchi occurs and can be observed on the routine chest films (Fig. 6A). Tomography will demonstrate this best but is not necessary nor is further investigation

via bronchography required. The area of narrowing occurs mainly at the level of the carina and in the majority of cases involves both main stem bronchi. The stenotic changes are progressive and eventually lead to severe respiratory difficulty.

Esophagus

The esophagus may be narrowed circumferentially (Fig. 6B). The appearance in this stage differs from the extrinsic compression caused by the nodes in the early phase. However, persistent nodular impression caused by residual lymph node enlargement is occasionally noticeable. The junction of the upper and middle third of the esophagus is still the area most frequently affected (Fig. 6B) although extensive narrowing is not unusual (Fig. 7B). The diseased esophagus is usually adjacent to the involved trachea and bronchi.

Superior Vena Cava Obstruction

Superior vena cava syndrome occurs frequently in this phase, although in our series this was encountered in only one patient. Fibrosing mediastinitis has been reported as probably the most common benign cause of superior vena cava syndrome. The superior vena cava is not only displaced but narrowed circumferentially (Fig. 8).

Pulmonary Vessels

Both the pulmonary arteries and veins eventually become encased by the enveloping desmoplastic tissue. The veins are morphologically thinner and more compressible than the arteries and are, therefore, affected earlier. Pulmonary congestion ensues, producing radiographic findings similar to mitral stenosis. Diffuse interstitial edema associated with Kerly A & B lines are observed. Chronic congestion eventually stimulates the production of fibrotic tissue which becomes superimposed upon the edema.

The pulmonary arteries are less frequently involved, but are nevertheless, eventually engulfed by the fibrous tissue. The vessel lumen gradually narrows and, despite the presence of pulmonary hypertension, becomes occluded.

Pulmonary arteriography was performed in three patients. The changes were striking. Severe stenosis of the branch vessels was encountered in one and total occlusion of the right pulmonary artery in the remaining two. In the one patient with incomplete occlusion, pin-point patency remained in the vessels supplying the right upper and middle lobes, as well

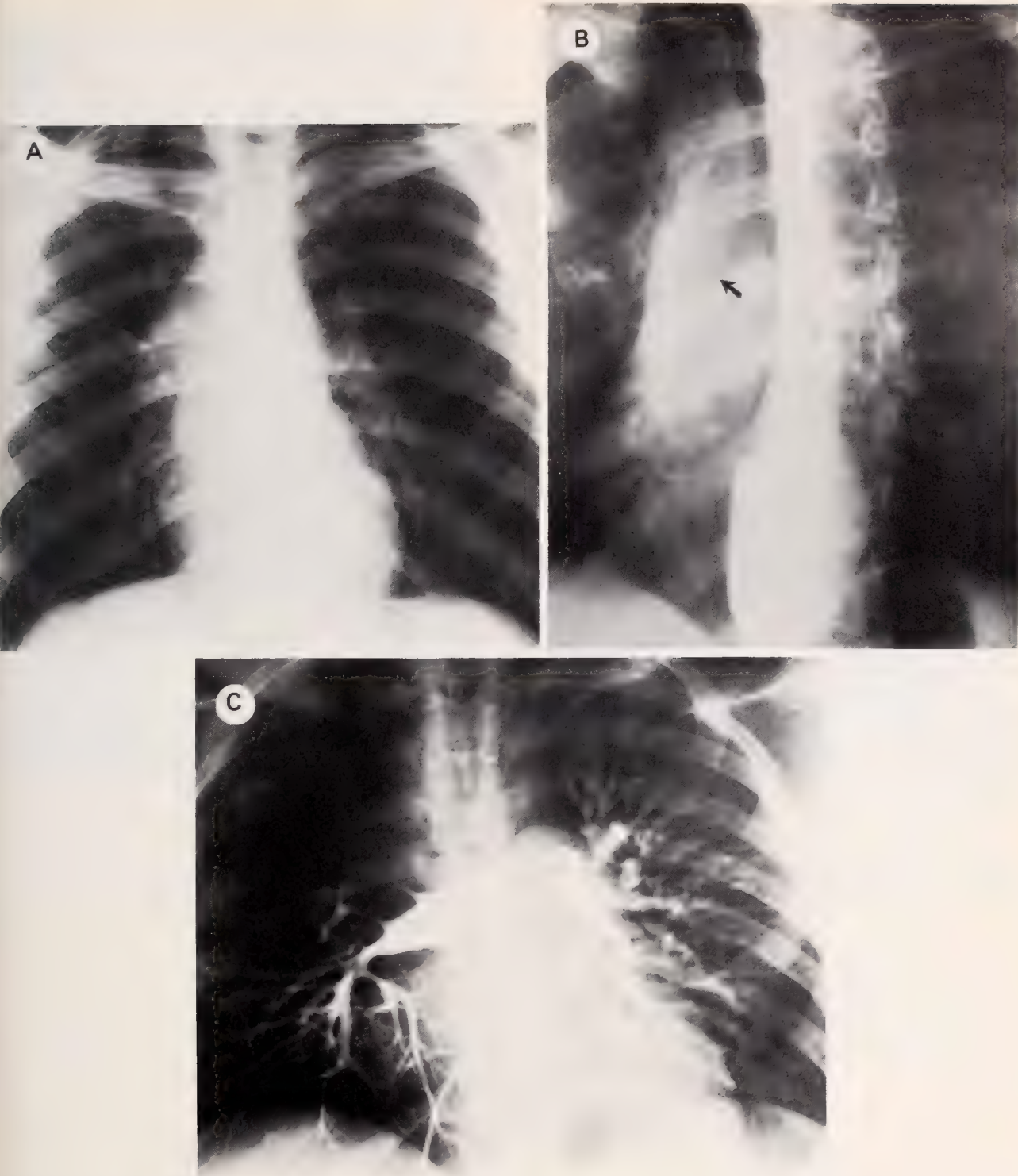


FIG. 6. Fibrosing Mediastinitis. Fig. 6A. The right side of the mediastinum is widened and irregular. The trachea is narrowed although poorly visualized. Fig. 6B. Concentric narrowing is present within the mid-portion of the esophagus and the narrowed trachea as well as the right main bronchus (arrow) are now well demonstrated. Fig. 6C. Pulmonary angiogram reveals only pin-point patency of the branches supplying the right upper lobe and some branches supplying the lower lobe. Note the difference in diffusion between the lung fields.

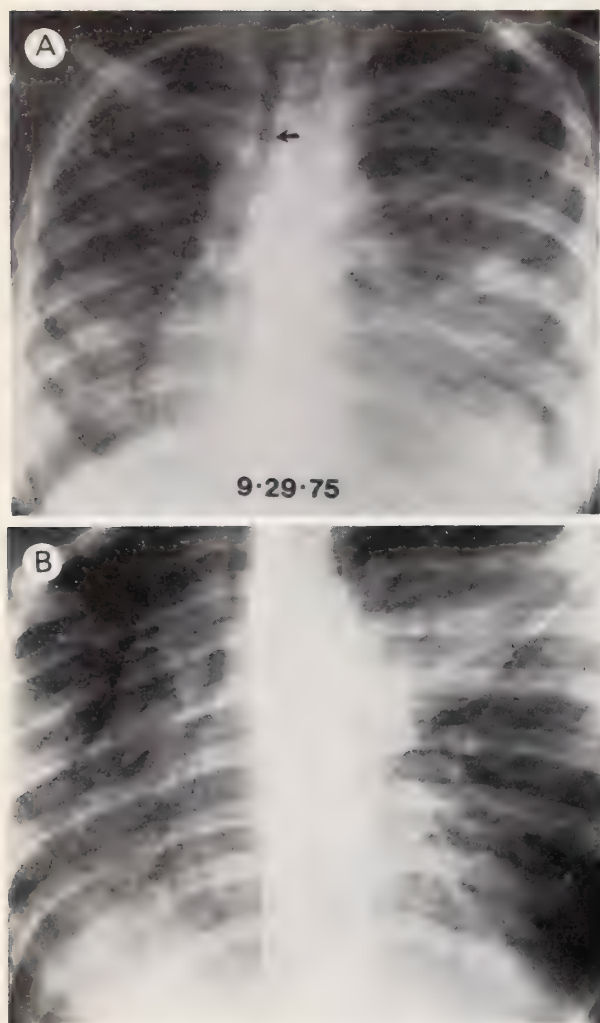


FIG. 7. Fibrosing Mediastinitis. Fig. 7A. Chest radiograph reveals a wide mediastinum with irregular borders. Diffuse infiltration is present throughout the lung fields due to congested vessels and interstitial fibrosis. The trachea is considerably narrowed. (Arrow). Fig. 7B. Esophagus shows diffuse narrowing.

as in some branches supplying the right lower lobe (Fig. 6C). Angiography in the other two revealed complete occlusion of the right pulmonary artery or its branches several centimeters after its origin from the main trunk but in close proximity to the nodal disease.

Discussion

Although the incidence of fibrous mediastinitis is far less frequent than mediastinal granuloma, the pathologic similarities that exist between the two abnormalities are so striking, that it is difficult not to conceive of them as representing varying stages in development of the same disease process. In

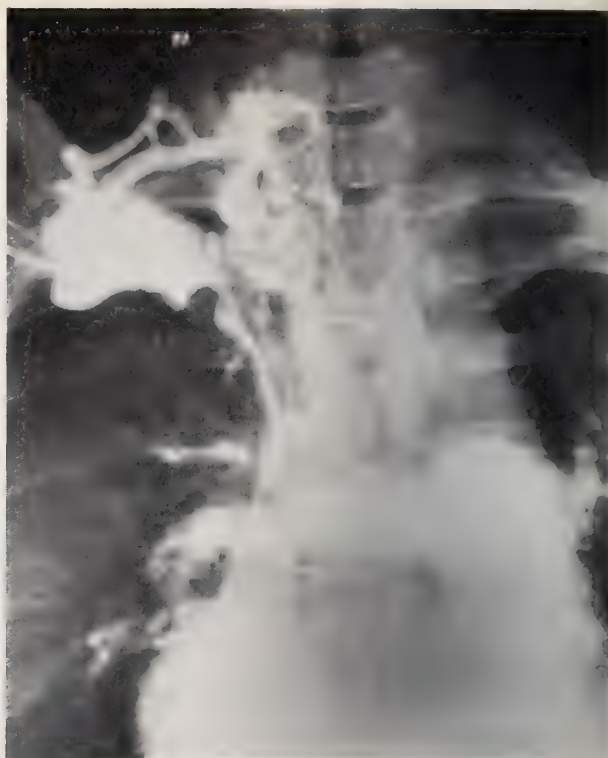


FIG. 8. Superior vena cava syndrome. Superior vena cava is displaced and narrowed. Vessels distal to the obstruction are distended and marked collateral circulation is apparent.

some cases, the histologic features of both blend into each other (5). Moreover, progressive radiographic evaluation of some patients revealed a definite progression from the early phase of nodal enlargement into the more chronic phase of fibrosing mediastinitis (5, 6).

Several explanations have been proposed to elucidate the mechanism of transition that occurs in these individuals. Spontaneous rupture of the nodes with spread of irritating contents throughout the mediastinum has been suggested but never demonstrated. Chronic lymphstasis due to venous and lymphatic obstruction with transudation of protein-enriched fluid and eventual fibrous formation is believed the cause of only a small percentage of cases. The most acceptable theory has been proposed by Goodwin et al (7). These authors have shown the fibrous tissue to initially deposit around the periphery of the infected node. This deposition is considered to be the result of a marked hypersensitivity response within the mediastinum that follows the seepage of an unidentified antigen into the surrounding tissue due to tissue breakdown within the nodes. As a result of this fibrous precipitation, the nodes seemingly increase in size (Fig. 9). A

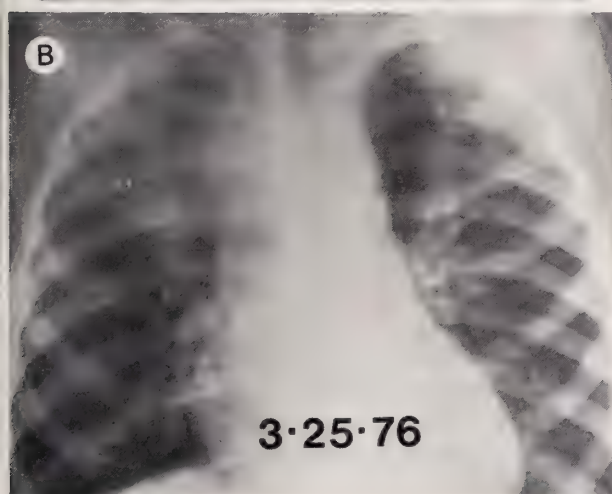
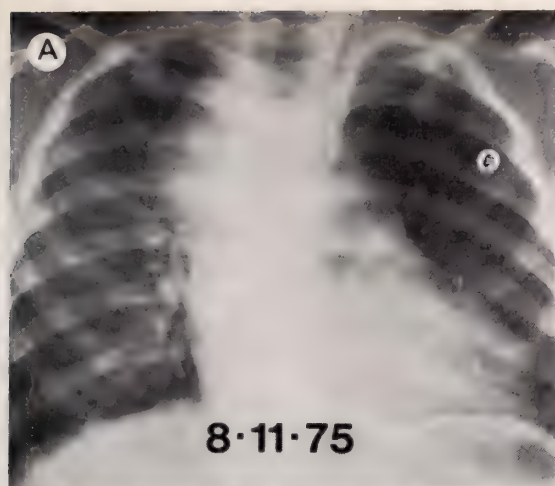


FIG. 9. Enlarging Lymph Node. 9A. The initial chest examination reveals large right-sided paratracheal nodes, with evidence of calcification. 9B. Several months later, the nodes have increased in size causing right upper lobe atelectasis. Diffuse mediastinal fibrosis was noted on mediastinoscopy.

similar mechanism is probably responsible for the observed growth of parenchymal histoplasmoses. The state of the host is therefore an important factor. Fibrosing mediastinitis is not common and seems to affect relatively young individuals with a strong immunologic response. Although tuberculosis has also been implicated as a cause of fibrous mediastinitis, the morphologic appearance of the nodes in both diseases is quite different. The nodes associated with histoplasmosis are engulfed and surrounded by dense fibrotic tissue, whereas in tuberculosis the nodes contain a relatively thin capsule and manifest only a minimal surrounding fibrotic reaction.

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Frequency of Neurologic Diagnoses: Implications for Curriculum Design

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Abstract

Neurologic diagnoses of inpatients at an urban primary care hospital and a tertiary care medical center were analyzed to determine the frequency and distribution of neurologic diagnoses and the neuroanatomic categories of these diagnoses. Diagnoses were recovered from the computer printouts of the Committee on Professional Hospital Activities-Professional Activity Study and data were compared for similarities and differences between the medical centers and the national and regional averages based on census divisions. After allowance for certain differences in types of neurologic material reported at different institutions, it was apparent that undergraduate and graduate training in diagnosis and management of neurologic diseases could be effectively planned by relating course or program content to the realities of hospital experience. Study of the differences between course content and the character of the clinical setting may also provide a basis for informed and constructive change in the design of medical school curricula.

Introduction

In 1975, it was estimated that 12 million people in the United States (U.S.) were afflicted with neurologic disease (1). Since most neurologic conditions are chronic in nature, involving long-term disability, the projected cost of medical care for these conditions may be greater than for many other categories of disease. Many neurologic problems (mental retardation, head injury, cerebrovascular disease, etc.) may never be seen by the neurologist or neurosurgeon. For many family practitioners, the medical school training in neurology may be the only opportunity to learn the signs, symptoms and management of neurologic disorders, especially those that are admitted as medical emergencies. Not all patients who have neurologic symptoms have primary neurologic disease. For example, peripheral neuropathy as a secondary manifestation of diabetes or systemic vascular disease must be recognized by the physician within the context of the patient's primary disease.

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Although clinical training of the physician must involve both ambulatory care and inpatient management, it is the latter that forms the basis for most of the initial phase of clinical training for the medical student. Effective clinical training requires an appropriately relevant education in preclinical sciences. It is therefore appropriate to examine the clinical problems present in a given hospital setting as a basis for planning the preclinical curriculum necessary to prepare medical students for dealing with these problems during their phase of clinical training.

The present study examines this approach for a single organ system (neurologic diseases) by analyzing 1) the frequency and distribution of neurologic diagnoses assigned to inpatients of an urban primary care and a tertiary care hospital center and 2) the neuroanatomic distribution of disease affecting these populations.

Materials and Methods

A retrospective survey was carried out in the medical record departments of City Hospital Center at Elmhurst, Queens (a primary care hospital) and The Mount Sinai Hospital of New York (a tertiary care hospital). Final primary and secondary neurologic discharge diagnoses were obtained from the Committee on Professional Hospital Activities—Professional Activity Study (CPHA-PAS) (2) com-

puter printouts in these locations during August, 1978. The diagnostic data in the CPHA data file are coded according to the International Classification of Disease (ICD) which lists diseases according to the systems affected as well as alphabetically. It was this handy, direct access to such specific and voluminous information that made a study of this type possible within a reasonable period of time.

For each neurologic disease listed in the ICD code, the total number of patients discharged with that diagnosis was computed. In similar fashion, the grand total of all neurologic diagnoses was determined for the three-year period of 1975–1977 for both Mount Sinai and Elmhurst Hospitals. From these figures, frequency of diseases, both by category and by anatomic distribution, was calculated. Data were then regrouped into 21 broad categories of neurologic disorders to match as closely as possible the available 1975–1976 CPHA-PAS categories of national and regional data based on census divisions. The national and regional data differ from those which are compiled at Mount Sinai and Elmhurst Hospitals in two respects: they exclude patients who died during hospitalization and/or whose stay exceeded 100 days.

Since neurology rotations for medical students at Mount Sinai School of Medicine take place both at Mount Sinai and Elmhurst Hospitals, the number of neurologic diagnoses for each of the 21 categories of neurologic diseases at these institutions were combined to truly reflect the total clinical exposure given to medical students and house staff. Each sum was then compared to the expected number of neurologic diagnoses for that category based on available PAS data for the Northeast region and the entire U.S. statistical interpretation of results was made on the basis of the chi-square goodness-of-fit test.

Limitations of the methods of this study include the following:

(a) All CPHA-PAS data excludes stillborns, patients with age, sex, date of admissions or date of discharge not recorded or misrecorded, patients with invalid final diagnosis codes or diagnosis/sex inconsistencies. Data are compiled from individual patient discharge abstracts submitted by hospitals participating in the Professional Activity Study (PAS) of the CPHA. The basic question of whether or not the information in the medical record is accurately reflected in the PAS case abstract must be kept in mind. An error in the patient's original method record, errors and/or omissions during the process of abstracting and processing information from the chart are all potential sources of inaccuracies.

(b) In 1975, 1848 hospitals participated in the Professional Activity Study while in 1976 the number was 1877 (2). In 1975, the number of short-term nonfederal hospitals which took part in PAS was 29.2 percent of such hospitals, while in 1976, it was 30.2 percent. In 1975, 56.7 percent of short-term nonfederal hospitals with 400–499 beds were represented in PAS yet only 23.4 percent of those with 50–99 beds were included. Similarly, in 1976, 55.2 percent of hospitals with 400–499 beds were represented in PAS but only 24 percent of those with 50–99 beds were included (2). Thus, relatively few of the smallest hospitals are included in the CPHA file. Because of the differences between CPHA hospitals and all accredited hospitals, the sample of patients for which data are available is not truly representative of all hospitalized patients.

Results

Neurologic diagnoses comprised approximately six percent of all diagnoses at Mount Sinai and Elmhurst Hospitals for each of the three years under study (1975–1977). Tables I and II list the frequency of neurologic final diagnoses by anatomic distribution at Mount Sinai and Elmhurst Hospitals for this period. It can be seen that diseases under the anatomic categories of cerebrum, thalamus and hypothalamus top the list, encompassing half the diagnoses at Mount Sinai Hospital and 80 percent of those at Elmhurst Hospital. Tables III and IV list the frequency of neurologic final diagnoses by disease category at Mount Sinai and Elmhurst Hospitals for the same period. Vascular diseases of the nervous system ranked first among all the neurologic diseases, accounting for about one-fourth and one-half of all neurologic diagnoses at Mount Sinai and Elmhurst Hospitals, respectively. Table V shows that the four census regions are not equally represented in terms of the percentage of hospitals participating in the PAS of the Commission on Professional Hospital Activities. The Northcentral region is over-represented and the South is under-represented.

Table VI presents the proportion of total CPHA patients in each of the four census regions having a primary and/or secondary neurologic diagnosis.

In this comparison, the Northcentral region has the greatest percentage of patients with neurologic disease. The relation between age and census region and the correlation between age and hospitalization for neurologic problems have not been analyzed, yet may partly account for this result.

Table VII shows a comparison of the frequency

TABLE I

Frequency of Neurologic Final Diagnoses by Anatomic Distribution, The Mount Sinai Hospital, 1975 to 1977 (abstracted from the H-ICDA coding of the Hospital Activity Report)

Frequency by Anatomic Distribution	No. of Diagnoses		
	1975	1976	1977
1. Skull, dura, leptomeninges, & site unknown			
leptomeningitis	62	60	38
subarachnoid hemorrhage	39	41	25
migraine	35	21	41
skull fracture	10	12	13
papilledema	10	9	10
phlebitis and thrombophlebitis of intracranial venous sinuses	1	3	—
subdural hematoma	1	2	—
SUBTOTAL (% of total diagnoses)	158 (7%)	148 (7%)	127 (6%)
2. Cerebrum, thalamus, hypothalamus			
acute but ill-defined cerebrovascular disease	245	178	144
cerebrovascular accident (stroke)	199	218	211
intracranial injury	133	70	130
neoplasm, metastatic	125	107	90
neoplasm, primary	10	62	57
convulsive disorder	58	39	30
other cerebral demyelinating disease	37	24	20
degenerative and atrophic disease	21	30	37
cerebral aneurysm	21	23	34
acquired hydrocephalus	21	38	29
congenital hydrocephalus	19	28	16
Down's syndrome	18	7	8
mental retardation	18	7	—
other spastic paralysis	16	5	13
transient ischemic attack	15	56	66
pseudotumor cerebri	12	5	15
cerebrovascular malformation	9	15	12
hypertensive encephalopathy	9	—	3
encephalocele	7	22	5
cerebral abscess	7	2	2
microcephaly	6	—	6
encephalitis	6	9	5
cerebral palsy	3	3	4
Amaurotic familial idiocy	2	2	—
glycogen storage disease	2	—	—
cerebral arteritis	1	—	2
multiple sclerosis	89	49	66
neurofibromatosis	—	12	12
cerebral edema	—	5	—
Wernicke's encephalopathy with brain syndrome	—	2	—
encephalomyelitis	—	1	3
lipid storage disease	—	—	1
SUBTOTAL (% of total diagnoses)	1109 (51%)	1030 (52%)	1021 (50%)
3. Basal ganglia			
Parkinsonism	233	181	192
other involuntary movement disorders	11	6	8
SUBTOTAL (% of total diagnoses)	244 (12%)	187 (9%)	200 (10%)
4. Brainstem, cerebellum, cranial nerve			
trigeminal neuralgia and neuritis	24	16	21
facial paralysis	—	20	27
Meniere's disease	24	14	8
neoplasm primary	21	15	10
neoplasm, metastatic	11	8	10
Horner's syndrome	—	7	3

TABLE I—continued

Frequency by Anatomic Distribution	No. of Diagnoses		
	1975	1976	1977
progressive bulbar palsy	1	1	—
hereditary cerebellar ataxia	1	2	—
other disease of cranial nerves	—	10	7
SUBTOTAL (% of total diagnoses)	92 (4%)	93 (5%)	86 (4%)
5. Spinal cord			
unspecified disease of the spinal cord	20	30	24
neoplasm, metastatic	13	11	8
spinal paralysis	11	13	1
amyotrophic lateral sclerosis	11	7	9
meningomyelocele	7	5	—
progressive spinal atrophy	3	15	19
myelitis	3	2	2
neoplasm, primary	2	5	—
hereditary spinal ataxia	—	3	2
syringomyelia	—	2	4
SUBTOTAL (% of total diagnoses)	74 (3%)	92 (5%)	79 (4%)
6. Peripheral nerve			
peripheral nerve disorder	210	216	187
spinal nerve neuritis and neuralgia	30	16	57
neoplasm, primary	25	32	34
SUBTOTAL (% of total diagnoses)	265 (12%)	264 (13%)	278 (14%)
7. Muscle			
myasthenia gravis	132	85	149
other myopathies	68	58	58
myotonic dystrophy	16	13	9
muscular atrophy	11	6	11
progressive muscular dystrophy	2	—	—
myositis	1	15	22
SUBTOTAL (% of total diagnoses)	230 (11%)	177 (9%)	249 (12%)
TOTAL NEUROLOGIC DIAGNOSES	2,172	1,991	2,040

of primary and secondary diagnoses of patients with neurologic disease at Mount Sinai and Elmhurst Hospitals with regional and national frequencies for 1976. Certain important differences are immediately apparent: Mount Sinai has a higher incidence of patients with malignant neoplasms, Parkinsonism, multiple sclerosis, and congenital anomalies of the nervous system than most other PAS hospitals throughout the country. Similarly, Elmhurst Hospital has a higher frequency of patients with epilepsy and cerebrovascular disease. The differences between Mount Sinai and Elmhurst Hospitals are all statistically significant ($p < 0.01$). There are a number of variables coming into play that may explain these differences. Among them are:

- differences between a voluntary teaching hospital and a city hospital in terms of patient population;
- differences in incidence and prevalence of disease according to age, sex, race, ethnic group,

etc.;

- differences in patterns of patient referral;
- differences in specialization of hospital staff and facilities.

The difference between the observed total number of neurologic diagnoses at Mount Sinai and Elmhurst Hospitals and the expected number of diagnoses based on Northeast and total U.S. frequencies is also statistically significant ($P < 0.01$). One may conclude that inpatient neurologic problems seen at Mount Sinai and Elmhurst Hospitals are not truly typical of those seen in other PAS hospitals. However, it must be stressed that hospitals participating in the CPHA-Professional Activity Study represent a minority of all short-term nonfederal hospitals. Furthermore, these hospitals are heterogeneous in terms of size, geographic distribution and patient population. Obviously, the greater the number of hospitals under the PAS medical records system, the greater would be the

TABLE II

Frequency of Neurologic Final Diagnoses by Anatomic Distribution, Elmhurst Hospital, 1975 to 1977 (abstracted from the H-ICDA coding of the Hospital Activity Report)

Frequency by Anatomic Distribution	No. of Diagnoses		
	1975	1976	1977
1. Skull, dura, leptomeninges, & site unknown			
subarachnoid hemorrhage	42	76	22
leptomeningitis	27	30	31
subdural hematoma	23	9	20
skull fracture	4	10	8
migraine	1	6	4
SUBTOTAL (% of total diagnoses)	97 (7%)	131 (9%)	85 (7%)
2. Cerebrum, thalamus, hypothalamus			
acute but ill-defined cerebrovascular disease	470	517	425
cerebrovascular accident (stroke)	51	123	115
neoplasm, metastatic	252	14	10
convulsive disorder	156	184	151
other spastic paralysis	43	—	—
transient ischemic attack	34	46	46
acquired hydrocephalus	12	15	10
Wernicke's encephalopathy with brain syndrome	11	19	17
Down's syndrome	10	4	9
multiple sclerosis	9	15	12
other cerebral demyelinating disease	4	4	1
neoplasm, primary	2	3	1
cerebral aneurysm	2	4	5
microcephaly	2	1	—
neurofibromatosis	2	1	—
cerebral abscess	2	2	2
congenital hydrocephalus	1	—	9
intracranial injury	—	212	187
cerebrovascular malformation	1	2	2
encephalitis	1	3	4
mental retardation	1	—	2
pseudotumor cerebri	1	—	3
cerebral edema	1	3	11
encephalomyelitis	1	—	—
encephalocele	—	1	—
degenerative and atrophic disease	—	1	—
other cerebral demyelinating disease	—	—	1
SUBTOTAL (% of total diagnoses)	1069 (80%)	1181 (79%)	1024 (80%)
3. Basal ganglia			
Parkinsonism	46	38	44
other involuntary movement disorders	3	3	2
SUBTOTAL (% of total diagnoses)	49 (4%)	41 (3%)	46 (4%)
4. Brainstem, cerebellum, cranial nerve			
facial paralysis	7	16	5
trigeminal neuralgia and neuritis	3	2	1
Horner's syndrome	3	—	1
other disease of cranial nerves	3	2	2
Meniere's disease	2	—	4
neoplasm, metastatic	2	2	3
neoplasm, primary	—	—	2
SUBTOTAL (% of total diagnoses)	20 (2%)	23 (1%)	18 (1%)
5. Spinal cord			
spinal paralysis	12	8	14
amyotrophic lateral sclerosis	4	4	5
meningomyelocele	4	—	1

TABLE II—continued

Frequency by Anatomic Distribution	No. of Diagnoses		
	1975	1976	1977
syringomyelia	1	3	—
myelitis	1	—	1
unspecified disease of the spinal cord	1	—	—
neoplasm, metastatic	—	2	—
neoplasm, primary	—	1	—
hematomyelia	—	2	—
progressive spinal atrophy	—	1	—
SUBTOTAL (% of total diagnoses)	23 (2%)	21 (1%)	21 (2%)
6. Peripheral nerve			
peripheral nerve disorder	29	61	8
spinal nerve neuritis and neuralgia	28	23	33
neoplasm, primary	2	—	3
neoplasm, metastatic	—	—	3
SUBTOTAL (% of total diagnoses)	59 (4%)	84 (6%)	47 (4%)
7. Muscle			
myotonic dystrophy	6	4	—
muscular atrophy	5	5	9
myasthenia gravis	2	3	5
other myopathies	1	2	9
myositis	—	3	3
SUBTOTAL (% of total diagnoses)	14 (1%)	17 (1%)	26 (2%)
TOTAL NEUROLOGIC DIAGNOSES	1331	1498	1267

TABLE III

Final Diagnoses by Disease Category, The Mount Sinai Hospital, 1975 to 1977 (abstracted from the H-ICDA coding of the Hospital Activity Report)

Frequency by Disease Category	1975 No. of Dx.	1975 % Total	1976 No. of Dx.	1976 % Total	1977 No. of Dx.	1977 % Total
1. Vascular diseases	540	25	539	26	498	24
2. Neoplastic diseases	207	10	251	13	231	11
3. Toxic-metabolic disorders	237	11	189	10	197	10
4. Muscle diseases	230	11	177	9	249	12
5. Other (other disease of cranial nerves, peripheral nerve disorder, cerebral edema)	210	10	231	12	207	11
6. Degenerative diseases	202	9	191	10	196	10
7. Traumatic and mechanical disorders	195	9	133	7	191	9
8. Infectious diseases	132	6	106	5	128	6
9. Episodic disorders	93	4	60	3	71	3
10. Symptomatic diagnoses (papilledema, eye movement disorders, Horner's syndrome etc.)	38	2	43	2	40	2
11. Developmental disorders	58	3	71	3	37	2
TOTAL NEUROLOGIC DIAGNOSES	2172		1991		2040	
TOTAL DIAGNOSES	35,063		35,594		36,347	
PERCENT NEUROLOGIC DIAGNOSES	6		6		6	

Dx = Diagnoses.

accuracy of these data on frequency of neurologic disease for any given area, whether it be local, regional or national.

Discussion

It should be pointed out that the data compiled for this study are derived exclusively from hospital

inpatient populations that have special features. They tend to contain a larger fraction of the rare or more severe diseases and may not reflect an accurate picture of the prevalence of disease. Therefore, findings obtained in a hospital population are not equivalent and cannot be directly extended to an outpatient population in a general practice setting.

TABLE IV

Frequency of Neurologic Final Diagnoses by Disease Category, Elmhurst Hospital, to 1977 (abstracted from the H-ICDA coding of the Hospital Activity Report)

Frequency by Disease Category	1975 No. of Dx.	1975 % Total	1976 No. of Dx.	1976 % Total	1977 No. of Dx.	1977 % Total
1. Vascular diseases	623	47	779	52	635	50
2. Neoplastic diseases	260	20	23	2	22	2
3. Episodic disorders	157	11	190	13	155	12
4. Infectious diseases	63	5	60	4	72	6
5. Traumatic and mechanical disorders	60	5	237	16	210	17
6. Toxic-metabolic disorders	55	4	57	4	57	4
7. Other (other disease of cranial nerves, peripheral nerve disorder, cerebral edema)	33	2	63	4	21	2
8. Degenerative diseases	36	3	44	3	38	3
9. Muscle diseases	14	1	17	1	26	2
10. Symptomatic diagnoses (papilledema, eye movement disorders, Horner's syndrome etc.)	11	1	19	1	8	1
11. Developmental disorders	17	1	9	<1	19	1
TOTAL NEUROLOGIC DIAGNOSES	1331		1498		1267	
TOTAL DIAGNOSES	21,635		21,409		21,628	
PERCENT NEUROLOGIC DIAGNOSES	6		7		6	

Dx = Diagnoses.

TABLE V

Region	1975 Total Short-term Nonfederal Hospitals	1975% in PAS	1976 Total Short-term Nonfederal Hospitals	1976% in PAS
Northeast	1027	31.8	327	32.2
Northcentral	1862	38.8	724	40.6
South	2332	20.5	440	21.0
West	1245	28.7	357	30.4

Source: Commission on Hospital and Professional Activities (CPHA) Professional Activity Study (CPHA-PAS) 1975, 1976.

TABLE VI

Percent Distribution of Patients with Primary and/or Secondary Neurologic Diagnosis by Census Region, 1975 and 1976

	Northeast	Northcentral	South	West	All U.S.
1975	22	40	20	18	535,138
1976	21	40	23	16	539,652

Source: Commission on Professional and Hospital Activities (CPHA) Professional Activity Study (CPHA-PAS) 1975, 1976.

A search of the recent literature reveals that very few publications have considered the possibility of using a profile or frequency of neurologic disease as a guide for designing undergraduate and graduate training programs. Most are concerned with defining the degree of knowledge of neurology that is essential to the graduating medical student, and the components of a suitable and meaningful graduate training program.

All medical students must be competent in assessing neurologic problems, especially those that are common, are acute emergencies, or are treatable (3). The question arises if emphasis should be

placed only on the neurologic diagnoses that are made frequently; that is, does "relevancy" mean that little or no time in a neurology curriculum should be spent on "rare" diseases, since the family practitioner may rarely encounter them? For example, although lept meningitis is relatively uncommon (accounting for approximately three percent of final neurologic discharge diagnoses at The Mount Sinai Hospital), every case should be diagnosed and treated early. As Murray (4) points out, the potential seriousness of a condition and the effect of intervention and therapeutic outcome, in addition to frequency, are the most important criteria that must be considered in making any diagnosis, whether or not it is neurologic.

It is generally agreed that the graduating medical student should have a firm grasp of the following (5, 6):

- assessment of whether or not the problem is in the nervous system;
- accurate anatomic localization of a lesion based on neurologic history and the patient's signs and symptoms;
- knowledge of the pathology, etiology, and appropriate therapy for the condition.

It is advocated by most authors that the internship, representing a transition year during which the medical school graduate gains experience in patient care, be retained as a requirement for specialization in neurology. This makes sense, since many neurologic disorders may be secondary to systemic disease and may not be confined uniquely to the nervous system.

TABLE VII

Percentage Distribution of Categories of Neurologic Disease: Primary and/or Secondary Diagnoses of Patients in CPHA Hospitals During 1976 by Census Region and Comparison with Mount Sinai and Elmhurst Hospitals in New York

Category	Northeast	Northcentral	South	West	All U.S.	Mt. Sinai Hospital	Elmhurst Hospital
1. Malignant neoplasm of brain	1	1	1	1	1	11	1
2. Malignant neoplasm of other parts of nervous system	1	1	1	1	1	1	1
3. Benign neoplasm of other parts of nervous system	1	1	1	1	1	2	0
4. Benign neoplasm of central nervous system	1	1	1	1	1	2	0
5. Inflammatory disease of central nervous system	1	1	2	2	2	2	4
6. Miscellaneous disease of central nervous system	7	9	10	9	9	6	1
7. Parkinsonism	2	2	2	1	2	11	3
8. Epilepsy	4	5	5	5	5	4	14
9. Subarachnoid hemorrhage	<1	<1	<1	<1	<1	2	6
10. Miscellaneous cerebrovascular disease	26	27	29	26	27	13	46
11. Precerebral occlusion	3	3	4	6	4	11	1
12. Aneurysm of head and neck	<1	<1	<1	<1	<1	1	<1
13. Peripheral nerve disorder except sciatica	10	14	14	16	13	12	4
14. Sciatica	1	2	1	1	1	2	0
15. Congenital anomaly of central nervous system	<1	<1	<1	<1	<1	4	<1
16. Skull vault fracture	2	2	2	2	2	1	1
17. Intracranial injury except concussion	4	4	4	5	4	1	14
18. Concussion	25	17	13	19	18	3	<1
19. Mental retardation	<1	<1	<1	<1	<1	<1	0
20. Transient ischemic attack	9	9	9	9	9	3	3
21. Multiple sclerosis	2	2	1	1	1	3	1
TOTAL	110,828	214,605	127,658	86,561	539,652	1703	1375

At this point it is worthwhile examining the usual content of a residency program in neurology (7, 8). Included are the following:

- experience in inpatient and outpatient care;
- emergency care of the unconscious patient; treatment of emergencies: coma, myasthenia gravis, status epilepticus, etc.;
- continuing care of the recently paralyzed patient including familiarity with rehabilitation measures used in the home;
- pediatric neurology: certain neurologic disorders are particular to childhood and their handling differs from that of adult neurologic disease;
- psychiatry: recognition and management of anxiety and other psychologic states;
- neuroradiology: performance, evaluation and recognition of indication for routine and special procedures, for example, x-ray, angiography, myelography, pneumoencephalography, CAT scan, brain scan, etc.;
- neuropathology: application of normal neuroanatomy and its alteration by disease; gross and microscopic examination of surgical and autopsy specimens;
- neuropharmacology and neurobiochemistry:

energy metabolism of brain, neurotransmitters, drugs: their indications, contraindications, side effects, toxic reactions.

The time devoted to these different components varies according to the institution.

The study by Rose (9) gives insight into the topics under discussion. A questionnaire was sent to 150 neurologists who had entered private practice within the past ten years to determine their opinions about the adequacy of their training programs. Of the 80 who responded, the consensus indicated that neurology residency programs should:

- place greater emphasis on clinical application of basic science (the need for using a problem solving approach to clinical cases rather than rote memorization of interviewing and examination methods was stressed. The rationale for this is self-evident: symptoms described and signs elicited will fit into a coherent pattern by progressive, orderly channeling of information rather than jumping to prefabricated conclusions.);
- keep the internship as a prerequisite for the neurology residency;
- provide more exposure to pediatric neurology, psychiatry and rehabilitation medicine since

these areas were perceived as weaker points in the respondents' training experience.

It should be possible to meet the learning objectives of both undergraduate and graduate neurologic training by considering the type of neurologic disease material available in the hospital setting and adjusting the educational program to take advantage of this material. Analysis of the annual inpatient data on diagnosis (for example, utilizing the CPHA-PAS data base) provides the first step for such educational planning. Similar data on ambulatory care might also be used for planning ambulatory care training.

Regrouping of the diagnostic data to identify categories of disease and the anatomic distribution of the diagnoses forms an obviously pragmatic basis for designing the anatomy, physiology and pathology components of a curriculum. The relative emphasis to be given to various topics within these disciplines can also be justified by this approach.

The study of a given curriculum from this point of view may disclose areas of over- or under-emphasis in the training program that are seemingly inconsistent with the clinical experience of the hospital setting in which training is taking place. Some discrepancies may reveal the need to restructure the curriculum to provide greater emphasis for appropriate topics. Others may be justified by the learning requirements of the program. The neurosciences course for first-year medical students at Mount Sinai School of Medicine, for example, devotes 15 percent of its hours to the study of spinal cord and peripheral nerve anatomy, physiology and pathology while only five percent of the neurologic inpatient diagnoses at The Mount Sinai Hospital

are concerned with this area of the nervous system. The sequence of the course, however, calls for early and detailed consideration of these topics since they form a model for later study of other regions of the nervous system.

Although the present report is concerned only with curriculum design for study of the nervous system, a similar approach could probably be used for examination of all other regional or specialized areas of undergraduate and graduate study in medical center training programs.

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Ampulla of Vater-A Misnomer

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Towards the end of the ductus choledochus, at its point of confluence with Wirsung's duct, there is a small area which, for over one hundred and twenty years, has been known as the "ampulla of Vater." This designation was accepted and rapidly came into general use among the majority of anatomists, with the opposition of but a few authors. Nevertheless, those few objections have not proved a deterrent to the now universally accepted definition.

Since gross errors have arisen in this connection, I decided to write a historical revision obtaining the original writings of all the principal anatomists who had considered the subject. Accurate translations had to be obtained before a final evaluation of the subject could be done.

The investigation was divided into three strictly chronological sequence: 1) from Andreas Vesalius to Giovanni Domenico Santorini and Abraham Vater, 2) from Abraham Vater to Claude Bernard, and 3) from Claude Bernard to Edward Boyden.

From Andreas Vesalius to Giovanni Domenico Santorini and Abraham Vater

Modern anatomy commences with Andreas Vesalius. His magnum opus *De Corporis Humani Fabrica* appeared in 1543 when Vesalius was only 28 years of age. It was truly a monument to science and was illustrated with beautiful plates drawn by Van Calcar (1).

Vesalius devoted volume V of his work to a description of the gallbladder and the biliary tree.

The precision of this description and the concepts therein are so clear that this work must be considered the starting point for all anatomists.

Vesalius was succeeded by his disciple Gabriele Fallopius (1523-1562).

Both died very young, but their names have persisted with everlasting fame. In his work *Obser-*



FIG. 1.
ABRAHAM VATER
1684-1751

vationes Anatomicae, Fallopius dealt fully with the biliary tract and became renowned for his five propositions regarding its operation, which gave rise to long and heated debates with Galenic physicians. Among the participants of these discussions, were the most renowned anatomists of that age, divided opinions being held by André Laurent (1550-1609), J. Rioloano (1577-1677), Thomas Bartholinus (1616-80), and his son Casper (1655-1738) (4).

Nevertheless, the most outstanding figure of this post-Vesalius era was Sir Francis Glisson (1597-1677) whose description of the biliary tract complemented the work of Vesalius. In his *Anatomia Hepatis*, Glisson gave an exact description of the site, constitution, and function of the sphincter of the terminal choledochus over two hundred years before Ruggero Oddi (9).

Exactly one hundred years elapsed before another decisive step was achieved by scientific de-

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velopment. Until that time and during the whole of the next century, both physicians and anatomists were unaware of the function of the pancreas and its true structure. It was a young and intelligent prosector of the renowned anatomist—John Vesling (1598–1649)—who took that step. That prosector was Johannes Georgious Wirsung (1600–1643).

In 1642, while carrying out a dissection, Wirsung discovered the main pancreatic duct. He communicated this in a letter addressed to his Master Rioloano (10). This discovery created great repercussion amongst medical circles, and seemed to be connected in some manner with Wirsung's tragic death in 1643. It was heatedly disputed since attempts were made to assign the discovery to Maurice Hoffmann, son of Casper Bartholinus, who was the bearer of the letter from Wirsung to Rioloano, dated July 7, 1643. A few days later (August 22, 1643) Wirsung was cowardly murdered. Wirsung had worked intensely conducting numerous dissections, in adult humans as well as in fetuses, dogs, cats, hogs, rats, frogs and hens.

Wirsung's discovery was soon disclosed and it enabled two other eminent investigators to confirm the secretory action of the pancreas. These investigators were Regnier de Graaf (1631–1673) (11) and Johann Conrad Brunner (1653–1727) (12). The former published his famous *Tractatus Anatomico-medicus de Succo Pancreatici. Natura et Uso* in which he demonstrated the true function of the pancreas, canalizing simultaneously in dogs the parotid gland and the pancreatic duct.

Brunner complemented De Graaf's studies and carried out the first partial resection of the pancreas in dogs, with a survival rate ranging between three months to one year. His book *Experimenta Nova Circa Pancreas* contains a description of his renowned experiment and in it he outlines the technique followed for his partial resection of the pancreas. He also demonstrated that ligating the duct did not affect the life of the experimental animals.

Page 286 of De Graaf's work contains an illustration in which a pancreas can be seen with dissected main duct and an open duodenum showing the orifice of the papilla, with the following description: "Communis ductus pancreatici et meatus biliaris quattuor circiter sub pyloro digitis transversis in intestinus exitus."

In explaining this illustration, De Graaf describes the characteristics of the main caruncle as we know it, pointing out, at the same time, the difficulties attendant upon the retrograde exploration thereof. Moreover, the illustration shows both ducts devoid of valves all along their course.

In 1670, Theodor Kerckring (1640–93) in his book *Observationes Anatomicae* described the valvulae conniventes in the duodenum (13).

In 1695, Godfridi Bidloo published in Amsterdam his magnificent work *Anatomie Humanis Corporis* (14) in which a plate appears of the pancreas and its ducts. At the duodenal opening there appears a ring of circular fibers, as previously mentioned by Glisson.

It is with this mass of knowledge that we arrive at the eighteenth century.

*Giovanni Domenico Santorini and
Abraham Vater*

One hundred and fifty years had elapsed since Vesalio's *Fabrica*. Slow progress had been achieved in anatomic knowledge as well as in the physiology of the terminal portion of the choledochus and its confluence with Wirsung's duct.

During the first quarter of the eighteenth century, two eminent investigators carried out anatomic studies and issued publications of findings and descriptions of such exactitude that they are still fully in force. I refer to Giovanni Domenico Santorini (1681–1737) and Abraham Vater (1684–1751).

Santorini was born in Venice, Italy, where he carried out all his scientific activities. Vater was born in Wittenberg, Germany, where he spent most of his life.

When only 22 years old, Santorini was appointed Public Professor of Anatomy. His lectures met with such success that this persisted until his death in 1737.

Santorini was an anatomist fully devoted to his Chair and to his studies. In 1724, he published in Venice his *Observationum Anatomicarum* and subsequently he continued working and illustrating his anatomic observations with magnificent plates drawn to precise scale. Santorini died in 1737 and his disciple Michael Girardi, Primary Professor of Anatomy edited his Master's work in Parma, 38 years after his death. In his book entitled *Anatomici Summi-Septemdecim Tabulae* one can admire the beautiful original illustrations drawn by Santorini as well as their magnificent descriptions.

Vater, who initiated his studies at Merseburg, obtained his Master's degree from Wittenberg University. He continued his studies in Leipzig and finally received his M.D. at Wittenberg University. Brilliancy attended his career as a teacher. In 1717, he was appointed Extraordinary Professor of Medicine at Wittenberg University; in 1719, he was transferred to the Chair of Anatomy and Botany to

which, in 1737, was added that of Pathology. In 1746, he changed for the Chair of Therapeutics which was considered the most important at the Faculty since it embodied the position of Dean.

Vater enjoyed a longer life, during the course of which he displayed an extraordinary academical activity. He was undoubtedly one of the physicians who enjoyed the greatest prestige in his days. At the age of 23, he lectured on the vital action. From then until his death in 1751, his scientific production was continuous and he was responsible for 112 publications. The wide variety of the subjects covered by Vater constitutes clear proof of his extraordinary skill. He was an accomplished anatomist and in his studies he injected wax into vessels and ducts, a technique he had learned from his Master, the renowned anatomist Frederick Tuysch. He studied such varied topics as the papillary extremities of the sensory nerves and described the sensitive corpuscles long before they became known as the Pacinian corpuscles. He also studied the salivary gland ducts. His activities covered such subjects as botanical catalogues, the action of certain medications, studies on canine rabies, a new method for variolar transplantation by means of incisions, several papers on biliary stones, and the manner of their formation. Vater's lecture on *New Biliary Diverticulum* was undoubtedly one of his least important works; however, it is the one which contributed most to making his name known to posterity (17).

Thus having described two parallel lives very briefly, this paper shall address the works of these authors that are intimately related to the subject matter of this investigation—the ampulla of Vater.

In paragraphs seven and eight of Santorini's book *Observationum Anatomicarum*, he deals with the duodenal intestine. The former paragraph is devoted to the form and nexus of the duodenal intestine (18). Herein, Santorini describes in great detail the location, the form, the size, the direction, and the external aspect of the duodenum. Paragraph VIII contains a full description of the internal aspect of the duodenum from the pylorus up to its terminal point in the jejunum. An admirable description is given of all the aspects of the duodenal mucosa from pylorus to jejunum with the Brunner glands, the Kerckring folds, both caruncles, and the "frenulum" which Santorini identified for the first time.

Santorini had completed these descriptions with illustrations precisely drawn to scale, the quality of which has never been equalled by any other anatomist. As regards the liver, the biliary tract and

the pancreas, Santorini prepared three illustrations (11–13). Each of these illustrations was accompanied by a careful description of the affected area which, as already stated, was drawn to scale with the respective measurements indicated at the margin so that one could establish the width and length of any structure. When Santorini died, the plates remained in possession of his disciple Girardi who was able to publish them in 1775.

It is interesting to transcribe a note added here by Girardi: "*Indeed, I always discovered the periform dilatation of the common duct, so-called choledochus, finding it sometimes greater, sometimes smaller, and in the proximity of the contracted mouth, not totally smooth but as if it were creased.*"

This is the first detailed description of the ampullary dilatation which is sometimes found in the terminal portion of the choledochus.

Abraham Vater's "Novum Bilis Diverticulum"

Dr. J. B. Blacke was kind enough to furnish me with a microfilm from the United States National Library of Medicine in Bethesda, Md., containing a complete copy of Vater's lecture, whose translation from the Latin was made available to us by Reverend Father José Ignacio Carranza (20).

Vater's publication reproduces the text of a lecture on anatomy—the fourth of a series—which he delivered at Wittenberg University on April 22, 1720.

As stated in the frontispiece, the lecturer was Abraham Vater, philosopher and physician, besides being Public Professor of Anatomy and Substitute Professor of Botany.

Vater's original publication consists of a title page, a preface and nine theses, and ends with an address to his respondent, Paulo Gottlob Berger.

For the purposes of the present study it is of interest to be aware of the complete translation of the title page, of theses seven and nine, and of the final address to the respondent (Fig. 2).

Anatomical Lecture 4—This lecture deals with a new bile diverticulum in the proximity of the opening of the choledochal duct, and a valvular process in the neck of the gallbladder. It also refers to the extraordinary utility of both structures in determining the bile flow.

The preface and the first six theses are devoted to the theme of the lecture and to describing the gallbladder and the biliary tract as well as the valvular arrangement of the cystic neck described by Heister as a spinal-shaped valve. Vater assigned its real value to this structure in the biliary circu-

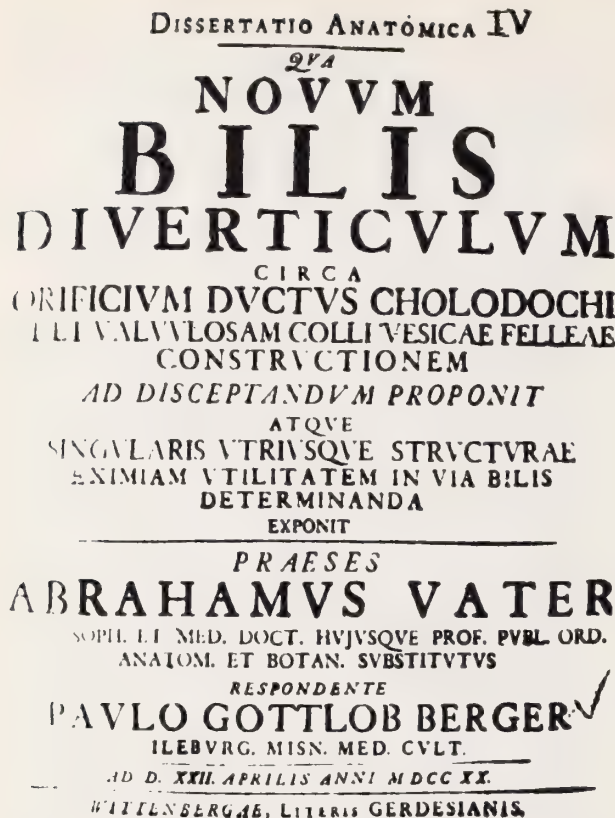


FIG. 2. Frontispiece of Abrahamus Vater's lecture, Wittenberg 1720.

lation and added comments and sensible deductions as to the role that bile plays in the process of digestion. The seventh thesis is devoted to the description of the diverticulum which he had discovered in the course of a postmortem examination.

Seventh Thesis

"But there still remains that which has motivated all these comments, viz: the singular structure of the duodenum, the common orifice of the pancreatic duct and of the choledochus which only recently we happened to discover. However, inasmuch as it can throw no little light in this matter we have deemed it worthy of being exposed to the erudite world.

"Having prepared an injection through the choledochus towards the intestine, and having ligated the opening thereof with the object of filling the pancreatic duct, our purpose was crowned with success. Indeed, the cereous matter penetrated not only into the excretory duct of the pancreas, but also around the opening and having surrounded it immediately gave rise to the formation of a little tumor, nevertheless uneven, sinuous, which we consider to be a deviation for the bile and the site where the mixture takes place with the pancreatic juice.

"It is true that we cannot affirm that the same arrangement is present in all the bodies inasmuch as we ourselves having again made the same injection in the same manner, the experiment did not yield the expected result because the little tumor did not appear; neither did the pancreatic duct become

filled. This, however, we believe should be attributed to the imminent even already present corruption in this cadaver, or even to the deficient ligature of the opening because if this is not carried out adequately the pancreatic duct can be easily entrapped and constricted by the material used for the ligature. However, in their preparation which for the reason given we have preserved, ocular inspection shows that the deviation is composed by the branches of the same pancreatic duct; or may be by the concurrent hepatic duct, especially in the external surface of the related intestine, where from all places can be seen the small vessels filled with wax. Neither does it seem to be far from the truth that such deviation is in the midst of the structure of the intestine in which the mixture takes place of the bile with the pancreatic juice and possibly also with the lymph from other small vessels, which mixture, on account of what has been previously stated, is indispensable before the bile is spilled into the intestine.

"What is truly certain is that said two ducts become united having regard to a more careful mixture of both humors, and not as described and delineated by all the anatomists, by a simple combination but by means of numerous ramifications. And this is confirmed by the renewed experiment as a result of which the insufflation of air into the common opening, we noted that here and there something became elevated within the internal membranes, and this we could disclose in the viscera of a child, preserved in liquid and still united among themselves.

"And even though this structure is not found in everyone; however, this observation is sufficiently singular and curious and is worth while being made known. The same observation may provide other anatomists with the chance of further investigating the same matter and of discovering many useful things in this respect. Inasmuch as Nature does not adhere in every case to the same order and a constant pattern, but frequently shows variations or departs entirely therefrom like we were able to ascertain in regard to the spiral valves which vary greatly in different individuals as taught by Heister, and in other individuals they are entirely lacking to the point of their existence having been denied by some or considered fictitious and imaginary by experienced anatomists.

"The same change we were able to discover recently in a cadaver of the female sex in which, upon attempting the injection previously referred to in order to disclose the deviation, this did not appear. However, the choledochal duct opened within the intestinal membranes, into two important branches with their separate openings into the intestine. But the corruption already present in this subject prevented a more careful investigation to the extent that there was neither the place nor the opportunity to examine adequately the numerous ramifications of these ducts which became dilated upon insufflation and were open to observation."

Ninth Thesis

"It has been our desire to describe these things briefly now, as allowed by the conditions of the weather and other matters, regarding the structure of the vessels which excrete bile, the arrangement of the canals which transport it, and particularly the spiral valves of the cystic duct and the deviation of the common opening into the duodenal intestine. We are forced to postpone a more ample description and delineation of this deviation until such time as having carried out many other experiments thereon we are in a position to examine the constitution in other subjects. Finally, it is only then when we will communicate it to the erudite world, developed and

represented by images, together with other curious anatomic observations. In fact, we do not fail to understand that a single observation is not sufficient proof. However, it is the Anatomist's obligation to examine carefully in the body each and every one of the things even the most insignificant, whose utility—though at times they cannot be described immediately—not rarely is apt to be very great.

"Praise and glory to God, for ever."

This ninth thesis ended Vater's lecture, and he listened immediately to the opposition of Paulo Gottlob Berger though it is not transcribed in the Wittenberg publication, which only contains the lecturer's reply, viz:

"The President extends his kind regards to the most noble and enlightened opponent, his beloved disciple.

"It is of the utmost importance that right from the start, he who devotes himself to the Art of Medicine should not only obtain the necessary scientific knowledge but also, through talks and erudite discussion, should exercise his intelligence to learn and express his thoughts, and to acquire self-confidence.

"In this manner he implants deeply into his memory everything he has learnt by hearing or reading; he gains that mental alertness which is so useful to discover the reasons for the different diseases; his intelligence becomes keener and thus he attains that eloquence which proves so useful to physicians.

"Indeed, upon attending the patient's bedside it will be necessary for him to pass a rigorous test, viz: explain to the patient the nature, the site, the causes and the course of his disease.

"He will be forced to banish the doubts and the objections of the friends of the patient, and to give an explanation to those who do not concur with his opinion. Thanks to that eloquence, the confidence of the sick and others present towards the physician is upheld and increased. For this reason it is not unusual for a loquacious physician to be considered a good physician by the ignorant.

"Now, being aware of your behavior, thanks to which you worry not only about learning the fundamental aspects of the Art of Medicine, but also to be in attendance giving obvious proof of your constancy, I cannot but extend to you my best wishes.

"I congratulate you, therefore, on account of your progress; I congratulate your illustrious father for such a son; I congratulate your fatherland; I congratulate myself on having such a student. I urge you, therefore, to continue progressing in your endeavor which is worthy of commendation. You may be sure that the most complete success will gloriously crown your achievements.

"Have a good time."

From G. D. Santorini and A. Vater to Claude Bernard

Some 136 years elapsed between Vater's lecture and Claude Bernard's famous *Mémoire sur le Pancréas*. This period is renowned for a personality whose prestige and scientific authority had an ample repercussion. I refer to Albrecht von Haller who was born in Berne in 1708 where he died in 1777 after having travelled all over Europe and having

dedicated himself to multiple activities. A member of 22 European Academies, Haller was a precocious genius. When only ten years of age, he composed poetry in Latin and in German. He obtained his medical degree at Leyden when he was 18-years-old. He was a disciple of Duvernois and above all of the renowned Boërhaave. From Holland, he travelled to England and France and then returned to his country whence he went to Hannover (Göttingen). Seventeen years later, Haller returned to Berne where he devoted himself to politics and to poetry, and wrote treatises on political economy. His scientific production was immense and has not been surpassed by any physician up to the present times. It covered numerous topics ranging from anatomy and physiology to clinical and surgical subjects. Moreover, he left a complete treatise on botany. His treatises on anatomy and physiology are extensive and monumental. In his famous *Bibliotheca* and *Disputations*, on anatomy, clinical and surgical physiology, he gathered together the most important European scientific works.

Vater's lecture was published in toto but without the address to the opponent, in *Anatomic Disputations*.

Concerning the terminal portion of the choledochus in his treatises on Anatomy and Physiology (21) Haller referred to this small area in the following words:

"Here it descends obliquely covered by a part of the gland (pancreas); it follows a course towards the inferior part of the second portion of the duodenum and becomes inserted from back to front some six inches from the pylorus in an oblong recess provoked by the pancreatic duct where it opens in a thinned orifice. Said recess runs along an ample course through the second cellular layer of the duodenum obliquely downwards; it perforates the nervous layer and runs again obliquely between this layer and the mucous layer; finally finding an opening in a long and protuberant duodenal wrinkle."

On page 433 of his 22nd book devoted to the pancreas, Haller describes a pelviform dilatation of the choledochus with a footnote reading: "Bertrandi, page 21 VATER de bilis divert" (Fig. 3).

In the course of our chronologic research, we arrive at the end of the eighteenth century without other important works relating to this area, with the exception of that of G.Th. Sommerring (1796) who introduced for the first time the word *Bauchspeicheldrüse* as the German definition for the pancreas as *abdominal salivary gland* (22).

Numerous works appeared in the first half of the last century relating to the terminal portion of the choledochus, the investigators dealing preferably with studies on comparative anatomy. Outstanding

which forms a sort of bile reservoir divided into several cells."

In J. F. Meckel's great treatise on Comparative Anatomy (French edition, 1823) he describes a large dilatation of the biliary tract in the duodenal tunica, in the following terms: "voluminous elongated dilatation, some three inches long, whose wrinkled internal wall gives birth to four transverse projections dividing this cavity into several small cells."

I have to point out that the elephant has no gallbladder and that this animal feeds exclusively on vegetables.

Cuvier, quoted by Gage, describes the existence, in cats, of a terminal portion of the "dilated choledochus between the tunica muscularis and the tunica mucosa of the duodenum, and their walls present several small cul-de-sacs which turn this cavity into an anfractuous structure. Thus, the pancreatic duct opens into the extremity of one of these cul-de-sacs."

All these descriptions coincide in reproducing the more or less ample image of the area we now know as the ampulla of Vater.

In his excellent study Tiedemann upholds the theory that the different arrangements of these ducts in animals are represented in the form of anomalies in human anatomy.

Claude Bernard and his "Mémoire sur le Pancréas"

In the winter of 1848, Bernard, who was studying the digestion of different alimentary substances, in the course of a comparative study between carnivorous and herbivorous animals, made a sensational discovery. He was experimenting with live dogs and rabbits and following the digestive process step by step. It was thus that he found that fatty substances became transformed as from the opening of the pancreatic ducts, which are differently situated in both animal species. Starting with these findings Bernard carried out exhaustive studies that were duly collected and published towards the end of 1856 in his famous *Mémoire sur le Pancréas*.

The extraordinary repercussion of Bernard's findings was accompanied by the diffusion of his Mémoire among the scientific circles throughout the world. Not only did Bernard present a study on the physiology of digestion, but he also reported his anatomic findings based on numerous dissections in human beings as well as different animal species.

Regarding the terminal portion of the choledochus and its confluence with the pancreatic duct, Bernard repeats the description contained in Santorini's *Septemdecim Tabulae*.

He gives a perfect description of the different varieties observed in humans, accompanied by numerous plates and schematic sections of the ampulla with the valvular formations described by Santorini, and the different varieties from the presence to the absence of the ampulla, and the separate exit of the two ducts in the duodenum (Figs. 5, 6).

When speaking about the ampullary dilatation of the terminal portion of the choledochus, Bernard states textually: "Avec lequel il s'abouche dans le fond d'une ampoule commune nommé ampoule de Water." A little further on he adds: "Les doux liquides ont du necessairement se meler dans l'ampoule de Water." Then, when mentioning an autopsy study on an individual who had suffered capital punishment, he repeats "L'ampoule de Water etait rempli de bile." In this same page he refers to "le pli de Water" and the surname Water is repeated four times.

In the pages containing the description of the illustrations of the ampulla, the name "Vater" is correctly written (Fig. 4).

Curiously enough, the mistake is not corrected in

(10)

les deux conduits est, du reste, parfaitement libre, et n'est gênée par la présence d'aucune valvule.

Les deux conduits pancréatiques se comportent différemment quant à leur mode d'abouchement dans le duodénum. Le petit canal récurrent, visible lors de la glande dans un trajet excessivement court, est parfois accompagné par le tissu glandulaire jusque sur la tunique musculaire de l'intestin, de sorte que le péritoine passe directement de l'intestin sur la portion du pancréas qui recouvre complètement le conduit. Après avoir traversé les membranes intestinales, ce conduit vient s'ouvrir sur une papille simple *g'* par un orifice étroit. Le canal direct, arrivé au niveau de l'intestin, subit un léger rétrécissement et traverse obliquement, de haut en bas, les tuniques intestinales en venant se placer au-dessous du conduit biliaire *V*, avec lequel il s'abouche dans le fond d'une ampoule commune, nommée *ampoule de Water*. Tantôt, comme cela se voit dans les *fig. 4* et *4 bis*, l'orifice du canal pancréatique *ij* s'ouvre dans le fond même de l'ampoule *a*, tout à côté de l'orifice biliaire *kk*; tantôt, comme cela se voit dans les *fig. 3* et *3 bis*, le conduit biliaire *k* se prolonge isolément jusqu'à l'orifice de l'ampoule. Dans ce dernier cas, la bile peut être apportée directement jusque dans la cavité intestinale sans se mélanger avec le suc pancréatique, tandis que, dans le premier cas, les deux liquides ont dû nécessairement se mêler dans l'ampoule de Water. On peut concevoir alors que si, par des circonstances accidentelles, l'écoulement de bile se fait avec abondance au moment où l'orifice de l'ampoule se trouve contracté, ce fluide pourra refluer dans le conduit pancréatique, surtout si celui-ci se trouve vide, comme cela a lieu pendant l'abstinence. C'est ce qui, sans doute, avait eu lieu chez un supplicié, que j'ai eu l'occasion d'examiner. L'ampoule de Water était remplie de bile, ainsi que le conduit pancréatique, qui était coloré en jaune, par cette sécrétion, dans plus de la moitié de sa longueur.

Outre la disposition dans l'abouchement des conduits pancréatiques que nous avons précédemment signalée, et qui est la plus ordinaire, on peut rencontrer un grand nombre de variétés, parmi lesquelles nous citerons les plus communes :

1°. Il arrive quelquefois que la disposition décrite plus haut est comme renversée, de telle sorte que le conduit principal s'ouvre le plus haut, et par une papille isolée, tandis que le petit conduit s'abouche en commun avec le canal choledoque (*fig. 2, Pl. 1-2*). Dans ce cas, l'ampoule, ainsi que le pli de Water, ne sont pas déplacés, et restent avec le canal biliaire et le conduit pancréatique inférieur.

2°. On trouve quelquefois les deux conduits pancréatiques à peu près

FIG. 4. Reproduction of page 10 of Claude Bernard's "Mémoire sur le Pancréas".

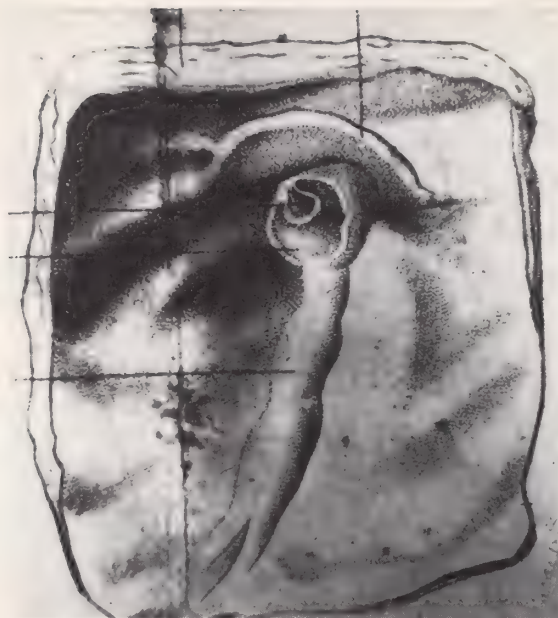


FIG. 5. Major Caruncle and "Frenulum"

the list of errata and throughout the paper no reference number appears corresponding to the works of Abraham Vater and Paulo Gottlob Berger.

From Claude Bernard to Edward Boyden

One hundred years elapsed after the famous *Mémoire sur le Pancréas* before the outstanding and fundamental achievements of Edward Boyden and his coworkers. An enormous quantity of papers, studies, books and treatises have dealt with this small area in the terminal portion of the choledochus at its point of confluence with Wirsung's duct and its duodenal outlet. I might mention that with very few exceptions, everyone has accepted the designation "ampulla of Vater" and this has been accepted universally as a firm designation. Consequently, I will not worry with all those who have accepted and continue using the designation, but with those who have raised doubts or criticized it. They are very few and I will mention them in the chronological order of this study.

Hubert von Luschka, Professor of Anatomy, Tübingen University, deals with the ampulla on two occasions. In 1863, in Chapter IV of *Die Anatomie des Menschen* dealing with *Die Bauchspeicheldrüse* upon describing it as "einen Weiteren Nische-Diverticulum Vateri" (27).

In 1869, he deals more extensively with it in a very complete study entitled *Die Pars Intestinalis des Gemeinsamen Gallenganges*. In chapter 3 entitled "The section of the choledochal duct, contained in the duodenal wall" (pages 94, 95) he studies in a



FIG. 6. In this cholangiography the characteristics of the "diverticulum," its union with the biliary tract and its partitions can be clearly appreciated.

detailed manner the area which, according to him, was described by "Berger unter Abraham Vater Abspizen," as "Diverticulum Bilis," a place where mixture takes place of the biliary and pancreatic secretions (28).

According to Luschka's comments, the work was performed by Berger under the auspices of Vater. Throughout the rest of Luschka's anatomic description the ampulla was always referred to as "Diverticulum Vateri." Only once more (page 96) does Luschka again place Berger in the first place attributing to him the error or designating it as "diverticulum" which is not found in *Omnibus Corporibus*.

In the bibliographic references, Luschka indicates that his access to Vater's work was through a transcription by Haller in his *Disputationes Anatomicae*, volume III, page 270.

G. P. Vlacovich (29), in 1893 (*Atti Real Istituto Veneto Di Scienze, Lettere, Arti*, pages 1185-1195, Volume 53, Sec. 7) published a paper entitled *Sulla Estremità Intestinale del Condotto Coledoco*. The only interesting point in this paper is a note reading: "Denominazione assolutamente impropria. Quanto ne disse il Vater fue ben poca cosa, no disgiunta da

grandi errori, che certamente non ne accrescono il valore."

Vlacovich, for his part, proposed giving the name of *Infundibulum ductus biliaris* to the portion of choledochus preceding its outlet into the ampulla. In no part of the paper is there any bibliographic reference to demonstrate that he had read Vater's original paper.

Oskar Stracker (30), in the first decade of the present century, in a very complete and documented paper entitled *Plica Longitudinalis* (Santonini's frenulum) devoted a whole chapter (II, page 399) to the "Diverticulum Vater" (Die Plica longitudinalis duodeni beim Menschen und bei Tierin, Bd. 118, Abt. 3, 1900, 375-437).

Stracker described the ampulla as we know it and in connection with the designation of *Diverticulum* utilized by Vater and Berger. He furnished an extensive note in which he transcribes Vater's phrase in Latin suggesting to other anatomists that they should complete his observation, and that in the second observation the instilled wax had expanded by the intestinal tunics of a cadaver in the process of putrefaction, thus suggesting unfavourable comments regarding Vater, his deficient technique, the cadaver undergoing decomposition, and the error in the term used, to all of which, he added Vlacovich's disparaging remarks.

After Stracker, no one else seemed to express interest again in Vater's lecture, to express doubt as to his paternity of the discovery, or to criticize his omission or possible mistakes. The designation of "ampulla of Vater" has been accepted and diffused universally.

A copious bibliography on the different aspects of its anatomy, physiology and pathology has accumulated since 1880 up to the present time, commencing with the magnificent studies by Simon Gage (31) and William F. Hendrickson (32), and continuing with those who have tackled the subject since 1900. Of all those mentioned there is no doubt that the series of communications, investigations and publications by Edward Boyden and coworkers from 1926 to 1960 constitute the most valuable contribution towards the knowledge of this small but complex area. Boyden's most important papers have been published in the time-honored journal *Anatomical Records* although the last and very complete paper regarding the anatomy of the choledochal-duodenal junction appeared in the June 1957 issue of *Surgery, Gynecology and Obstetrics*. Of all these publications, the one which has seemed to us particularly useful from the standpoint of this investigation is the one entitled *The Pars Intestin-*

alis of the Common Bile Duct as viewed by the Older Anatomists (Vesalius, Glisson, Bianchi, Vater, Santorini, Haller, etc.) (*Anat. Rec.*, 66:217, 1936).

A scientific error that has persisted for over a century

A careful reading of all the studies carried out of the terminal portion of the choledochus over the past two hundred and fifty years since the lecture delivered by Abraham Vater at Wittenberg in 1720, has enabled us, we believe, to elucidate a scientific error that has persisted until these days. This error consists in assigning the name of Vater to the ampullary dilatation preceding the ostium of the major duodenal caruncle.

In order to clear up the problem it is indispensable to find an answer to the following queries:

1. Who was the first anatomist to describe this ampullary dilatation?
2. Who was the first to give the name of "ampulla of Vater" to said area?
3. What did Abraham Vater describe?

The first description of the ampulla was beyond all doubt, that of Giovanni Domenico Santorini. Before his time, all the anatomists referred to the confluence of the two ducts as a simple union. Upon describing "Tabula XII" of his book *Septemdecim Tabulae* edited by Girardi in 1775, Santorini gave a very precise description of that vesicular dilatation, that can be observed in some healthy and robust subjects.

After Santorini, it was Albrecht Haller who described an "oblong sinus" whence the biliary and the pancreatic ducts open up.

Between Haller's and Claude Bernard's times, the anatomists devoted to the study of comparative anatomy, especially Cuvier and Meckel, described the ampullary dilatation in animals devoid of gallbladder, particularly noteworthy in elephants.

Tiedemann found such arrangement in animals, and described several anomalies in humans considering that these were representative of the findings in other animal species.

It is noteworthy that none of these investigators associated the existence of the ampullary dilatation with the name of Abraham Vater or that of Gottlob Berger.

In accordance with Haller's reference, I instituted a search for the book by Amerogio Bertrandi whose investigations in anatomy found ample diffusion in Europe between 1780 and 1850. I was fortunate enough in being able to consult the 1786 edition (published in Latin, in Torino, Italy) in the Faculty

of Medicine of Buenos Aires (Fig. 7). On pages 153 and 154 of this book, Bertrand describes the cholechochal duct and its opening into the duodenum after penetrating, together with the pancreatic duct, into an oblong cavity. A footnote contains a long bibliographic quotation that describes the terminal dilatation in relation to the lecture by Abraham Vater on the diverticulum (Fig. 8) (37).

It is from this bibliographic quotation that the equivocal situation arises regarding the real situation of this "diverticulum" described by Vater.

On the basis of all our research, we arrive at the conclusion that it was Claude Bernard who, for the first time, referred to the "ampulla of Vater" in some parts and to "De Vater" in others.

It is really curious that all the publications preceding *Mémoire sur le Pancréas* do not mention

Vater anywhere. Instead, Vater's name appears in all subsequent publications. I have consulted anatomy treatises published before Bernard's *Mémoire*, especially Cruveilhier's first two editions (1837 and 1845). Not only does Vater's name not appear therein, but the description makes no reference whatever to the existence of the ampulla.

Instead, in the fourth edition of this treatise (dated Paris, 1865) the ampulla appears with Vater's name and there is a reproduction of one of the illustrations by Claude Bernard (34).

OPERE

DI

AMBROGIO BERTRANDI

PROFESSORE DI CHIRURGIA PRATICA NELLA R. UNIVERSITÀ
DI TORINO, MEMBRO DELLA REALE ACCADEMIA DI
CHIRURGIA DI PARIGI, DELLA SOCIETÀ REALE DI
TORINO, E PRIMO CHIRURGO DELLA S. R. M.
DEL FU RE CARLO EMANUELE

PUBBLICATE, E ACCRESCIUTE DI NOTE, E DI SUPPLEMENTI

DAI CHIRURGHI

GIO. ANTONIO PENCHIENATI

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DI TORINO

TOMO I.



TORINO MDCCLXXXVI.

PRESSO I FRATELLI REYCENDS.

Con Privilegio di S. S. R. M.

FIG. 7. Frontispiece of the "OPERE" of Ambrogio Bertrandi. Torino 1786.

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H E P A T I S

fini propior. Ab his continuatus *choledocus duodenus* pone primum duodeni arcum descendit, subitque pancreatis posteriorem partem illi intestino adnexam, inferiturque posteriori faciei inde dux secundæ partis duodeni, &, obliquo ductu ejus tunicas penetrando, in ejusdem cavum aperitur (a). XIX.

(a) Osculo oblongo, superius arcuato, inferius acuto, adinstar denticulipri Winslow p. 533. Edit. Paris. in H., quam ut plurimum adinstar fissuræ oblongæ ani corrugatam observavimus, circumductis Peyer's villis, qui porro ad aliquam distantiam rugosi producuntur, & sensim inde evanescent, eosque *frenula*, aut *ligamenta* appellavit Santorinus observat. Anat. cap. 9. Sat bene expressam carunculam non vidimus, quam papillarem eminentiam vocat Blanchardus cap. 23. part. 2. Anat. Reformat., aut laxam quamdam membranofam eminentiam Muniks. Anat. Nov. p. 44. vel duris membranulis, utrinque ad orificii latus laxè prorepentes, quas describit Vesalius l. 5. c. 8. Nonnulli vero scribunt, illic intimis intestini tunicis corrugari, implicarique in modum valvulæ deorsum penditis, & *clitoridis præputium* quodam modo referentis. Memoratas autem rugas adeo satis laxas, ut hanc comparisonem admitterent, vix in uno homine dissenteria denato vidimus. Joannes Muraltus Epist. 5. ad Peyerum, circum hoc orificium in Bove, plexus duos glandulosos notavit longitudine pollicem æquantes, qui quidem minores in Homine occurrunt. Cum *Choledeco ductu* aliquando etiam *pancreaticus* aperitur, unius vero osculum ab alterius osculo per dimidiam unciam distans vidit Brunnerus Exper. Nov. Pancreat., ad minorem distantiam alii, semper vero aliquam idem Brunnerus, quod omnino falsum in Homine. Ductum *choledocum* duplicem in intestinum duodenum describit Falloppius l. c., eorum alterum in ventriculum Vesalius ex Pontificis trementum Remige lib. 5. cap. 8. pag. 436. Edit. Boeh., & Aibin.

Abraham Vaterus Dissert. de Diverticulo bilis amplitudinem quandam eo in loco, ubi hec vasa in intestinum duodenum aperiuntur, de cæcib. Vid. Kalm. Tab. Anat., nec quidem aliud esse videtur *sinus novus* in hac parte a Bartholino descriptus pag. 79. Anat. Reformat., perpetuus Brunnerus l. c., non perpetuus ipsi Vatero l. c., & solum in Ascite visus a Peyer's Patolog.

FIG. 8. Page 154 of the "Opere" of Ambrogio Bertrandi, with the reference to "Vater's Diverticulum."

It appears that all the French anatomists subsequent to the publication of Bernard's *Mémoire* reproduced his illustrations.

Whence did Claude Bernard obtain the information that such area was named "ampulla of Water?" The error in the text may have been a printer's mistake but the author did not correct it in the list of errata and allowed doubt to persist as to whether the true surname was Vater or Water. On the other hand, we must presume that Claude Bernard did not read Vater's lecture, nor its reproduction in Haller's compilation. What did Vater describe?

The answer to this query does not offer any difficulty when considering our present knowledge. There is no doubt that Vater described what we now denominate perivaterian diverticula described by Le Tulle (35) in 1899. The duodenal diverticula were scarcely known and the first mention of this anomaly was published in *Histoire de l'Académie Royale des Sciences* (Paris, 1710, page 37). The second mention corresponds to Morgagni in 1761. Another observation was described by Rahn in 1796. Apparently all these cases were diverticula of the first portion, in the proximity of the pylorus.

Consequently, it is no wonder that Vater or Claude Bernard, were not aware of these formations which were described towards the end of the century by Le Tulle, and at the beginning of the twentieth century by Baldwin (36). With the advent of radiologic studies, the diverticula commenced to be described with greater frequency after 1913. It should be noted that in Baldwin's series, they were present in 14.2% of the postmortem studies (36).

I have been able to study a quantity of perivaterian diverticula, and their characteristics are fully coincidental with the description passed down to us by Abraham Vater (Fig. 9, 10). Radiographs are reproduced of two of these cases in which can be appreciated that the simple ligature of the terminal portion of the papilla is sufficient for the little tumor described by Vater to make its appearance.

Consequently, all the criticisms advanced by Luschka, Vlacovich and Stracker can be considered unjust and entirely groundless. I must say the same regarding the authorship of the lecture.

No doubts should be entertained as to Vater's paternity thereof. Paulo Gottlob Berger was one of Vater's disciples and acted as his opponent in a sort

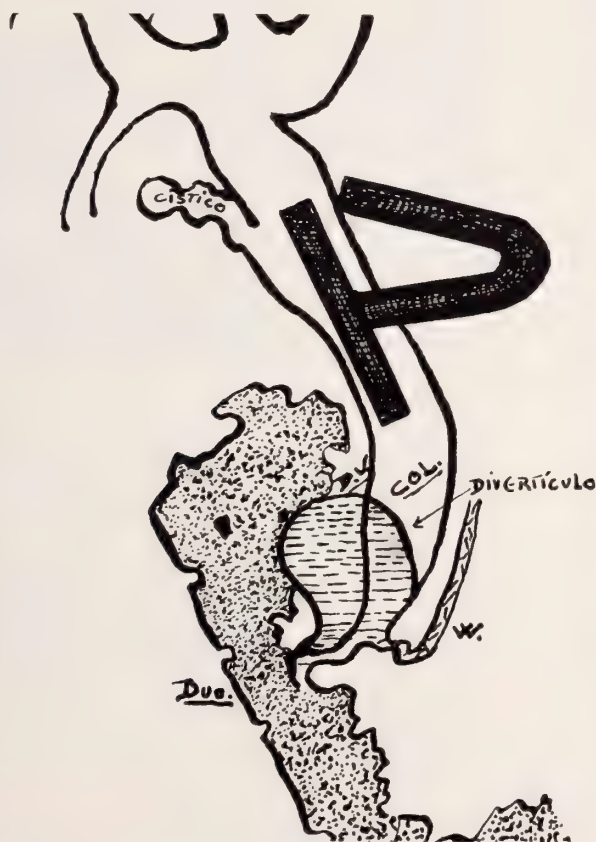


FIG. 9. Vater's diverticulum (personal observation). Postoperative serial cholangiography in a case of choledocholithiasis.



FIG. 10. X-ray of Wirsung's duct in anatomical specimen of duodene-pancreas (personal observation). Ampulla with Santorini's valves and accessory duct.

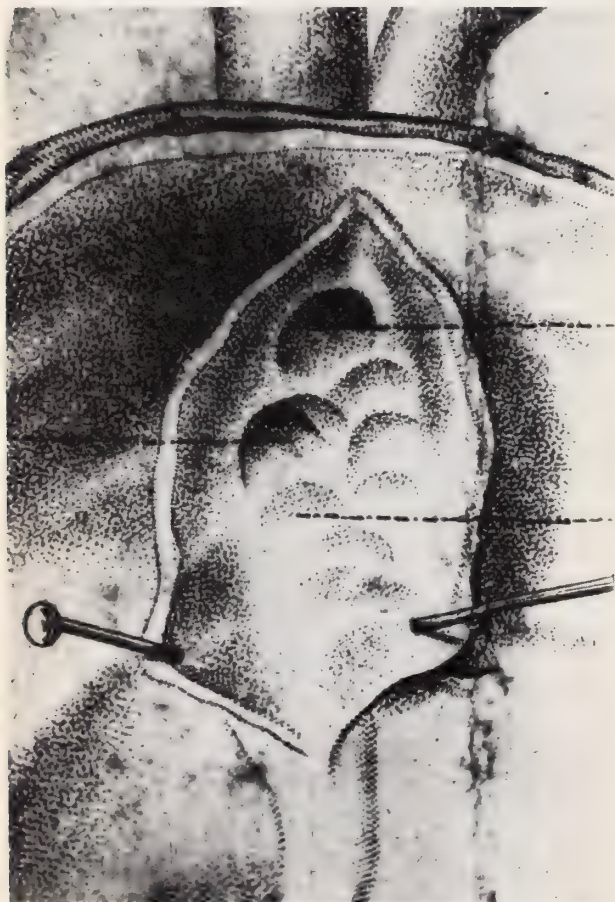


FIG. 11. Ampulla of Vater (open in sagittal section) showing Santorini's valves. (These figures correspond to figures 4 and 4 bismplate I, of "Mémoire sur le Pancréas").

of academic competition. It is unexplainable that this equivocal situation should have been maintained on the part of German authors.

As far as Claude Bernard is concerned, the error is excusable because the finality of his work was



Fig. 3 bis



FIGS. 12-13. Major caruncle and its sagittal section. These illustrations correspond to fig. 3 and 3 bis of plate I, of "Mémoire sur le Pancréas".

different. The anatomic study carried out by him and the pertinent illustrations (Figs. 11-13), constituted a valuable contribution towards the knowledge of the area in question.

I have made a careful investigation of Claude Bernard's monumental work with the intention of finding some explanation to this error, but upon commenting on the choledochus and Wirsung's duct he made no mention whatever of Vater nor his diverticulum. It is highly probable that Bernard might have read Ambrogio Bertrandi's book and thus drawn this erroneous information.

The extraordinary world-wide diffusion of *Mémoire sur le Pancréas* explains the ready acceptance of the designation "ampulla of Vater" given to a structure that was not described by this author.

Notwithstanding the years that have elapsed, I am convinced that the error should be corrected. The name of Vater applied to the dilatation described by Santorini, besides being erroneous, constitutes an injustice. For my part, I propose that this area be named after its discoverer. In other words, that it be named "ampulla of Santorini." Vater should be honored by designating "Vater's diverticula" the misnamed perivaterian diverticula. In this manner, each author would be assigned the merit due to him.

Acknowledgments

This work was made possible by the collaboration of the following people who were kind enough to enable me to consult the original books, and to obtain innumerable photocopies of papers and their illustrations. I am also thankful for the translations from the Latin. These people are: Dr. Horacio Hernández, Director, Library of the Faculty of Medicine, Buenos Aires University; Dr. John B. Blacke, Director, History of Medicine Section of the United States National Library of Medicine, Bethesda, Md.; Reverend Father J. Ignacio Carranza, erudite Latinist; and Dr. J. De Vega from the Gerardo Ramón Library who, with his continuous and invaluable help, made available to me the scientific works of the last centuries.

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Pleural Peritoneal Mesothelioma: A Case Report

BURTON COHEN, M.D. AND STAVROS EFREMIDIS, M.D.

Although mesothelioma of the omentum is a rare lesion, the angiographic findings are helpful. Ultrasound, and computerized tomography are also useful in ascertaining the character and extent of the lesion.

Case Report

The patient was a 67-year-old white male, retired hotel manager. He presented with left rib pain, a left axillary mass, and a large left upper quadrant mass. Roentgen examinations prior to admission demonstrated lytic lesions of the left fourth, fifth, sixth, and seventh ribs. Extrapleural soft tissue densities were noted in association with the rib lesions. A bone scan was positive in the same area. A liver spleen scan, intravenous urogram, barium enema and upper gastrointestinal series were all negative except for displacement of normal structures by a left upper quadrant mass. Following these studies, the patient was admitted for further evaluation. Ultrasound was performed, and demonstrated an irregular mass, anteriorly located in the left upper abdomen (Fig. 1a, 1b). The mass had a mixed echo pattern, with solid and cystic components. The liver, spleen and kidney were within normal limits.

Computerized tomography (CT) confirmed the sonographic findings (Fig. 2). Celiac and superior mesenteric arteriograms demonstrated an otherwise normal pancreas and spleen displaced cephalad (Fig. 3a, 3b). Downward displacement of the distal transverse colon was also noted, as was medial

displacement of the gastroepiploic artery by a mass. The gastroepiploic artery appeared to be draped over the mass. Hypertrophied omental branches with accompanying tumor stain were noted, which characterized the lesion as a malignant tumor of the omentum. A biopsy of the left axillary mass was performed. It demonstrated a lymph node completely replaced by what was initially thought to be an anaplastic carcinoma with a marked desmoplastic reaction.

A laparotomy was performed and a 12" × 10" cystic necrotic omental mass was found. The colon and stomach were both adherent to the mass. Pathologic study demonstrated that the omentum was infiltrated by mesothelioma. A segment of colon showed infiltration of the serosa and omentum by highly anaplastic mesothelioma. Subsequent comparison with the original lymph node biopsy demonstrated an identical histologic pattern of mesothelioma.

Discussion

CT and ultrasound demonstrated a mass anteriorly in the left upper abdomen. This location is compatible with a lesion in a variety of structures including the tail of the pancreas, stomach, and anterior abdominal wall, as well as the omentum. Angiography was definite in the localization and characterization of the lesion. Hypertrophied omental vessels are quite diagnostic of malignant omental tumor. According to Deutsch et al in a review of 350 cases, hypertrophied omental vessels were only seen in the cases with malignant omental involvement (1). Stretching of omental vessels as well as a tumor stain are often seen in omental mesothelioma (2).

Mesotheliomas more commonly involve the pleura rather than the peritoneum. Primary tumors of the peritoneum are extremely rare (3, 5, 6).

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FIG. 1. Transverse (a) and longitudinal (b) sections of an abdominal ultrasound study show a large irregular ecogenic mass (M) in the upper abdomen anteriorly.

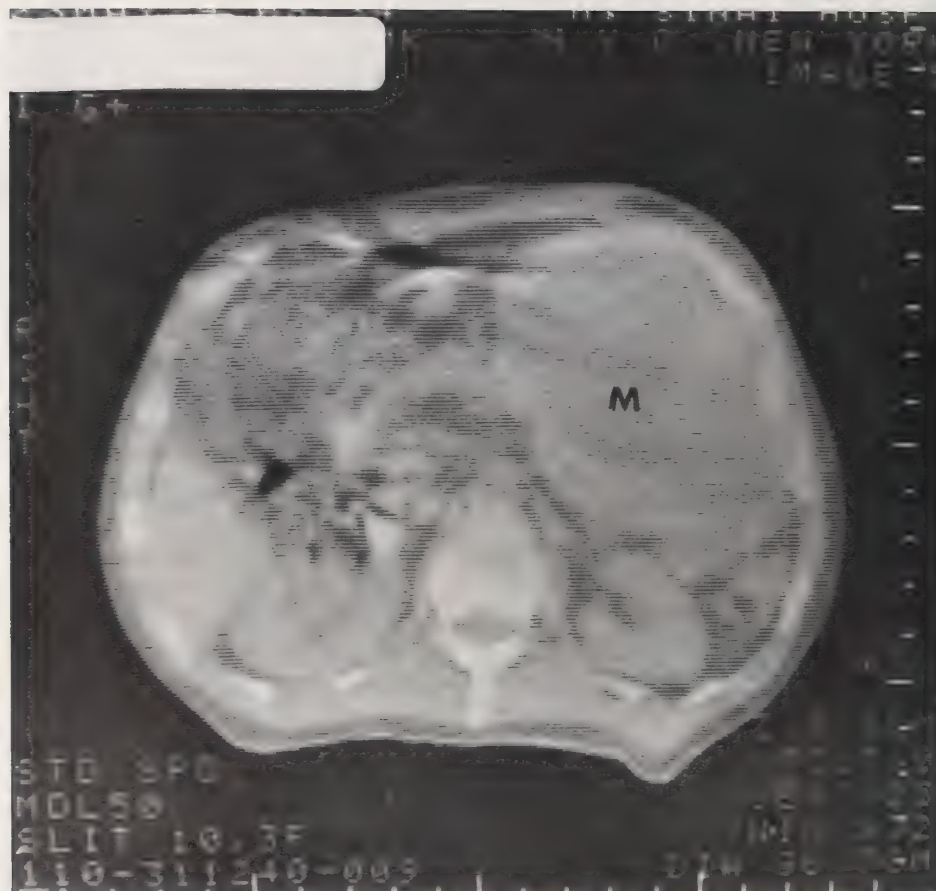


FIG. 2. Computerized tomography of the abdomen at the level of the renal sinuses shows a large, homogenous and well demarcated peritoneal mass (M) on the left side of the abdomen.



FIG. 3A. Arterial phase of a celiac arteriogram. Abnormal vascularity (arrows) from omental vessels coming off the left gastroepiploic artery. 3B. Venous phase of the same study shows tumor staining (arrows) at the same area.

Occasionally, mesothelioma involves both sides of the diaphragm. There is an increased association of both pleural and peritoneal forms of the disease with asbestos exposure. It has been found that patients with peritoneal tumors did not differ in their type of exposure from patients with pleural tumors (4). Although ultrasound and computerized tomography are helpful in assessing the extent of the mass, angiography is superior in definitely localizing the mass in the omentum, and in characterizing it as malignant.

Acknowledgment

We are grateful to Dr. Harold Mitty for his assistance and guidance.

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Unusual Problems in Surgery

A. Robert Beck, M.D., Editor

Partial Splenectomy in a Child

LARRY E. SHINDELMAN, M.D., LEONARD KASEN, M.D., AND A. ROBERT BECK, M.D.

Debate over the management of the injured spleen in children began in 1971 when Ein, Douglas and Simpson (1, 2) presented their paper on non-operative management of splenic trauma. In part, the impetus for alternatives to splenectomy stemmed from King and Schumaker's report of overwhelming sepsis in five children after splenectomy for hereditary spherocytosis (3). This non-operative approach received additional credence when Singer (4) in 1973 extensively reviewed the subject, and found that splenectomized children had a 50% greater incidence of sepsis, and a 50% greater fatality rate from sepsis as compared to children with intact spleens. This debate later assumed a third constituent when Mishalany (5) published in 1974 the first report of splenic repair in an eight-year-old boy. Since that first report by Mishalany, numerous investigators have reported on successful splenorrhaphy in children with splenic trauma (6-12).

The purpose of this paper is to present a case of serious splenic rupture that was successfully repaired, and to focus on the current debate in the management of splenic trauma that is presently being discussed in the literature and at our institution.

Case Report

L. M. is a ten-year-old boy who was admitted to The Mount Sinai Hospital with complaints of left

upper quadrant pain, left shoulder pain, and occasional dizziness on standing following a sledding accident. On physical examination, the patient was conscious, pale and apprehensive. His blood pressure was 98/70 mm Hg, heart rate 107 beats/minute. The pertinent physical findings were limited to his abdomen, demonstrating left upper quadrant tenderness and guarding. Laboratory examination revealed a Hgb of 11.3 mg%, Hct 32%, WBC 21,000 with a shift to the left. The urinalysis, amylase and serum electrolytes were all within normal limits. Roentgenograms showed no evidence of rib fracture, well expanded lung fields bilaterally and unremarkable abdominal gas pattern. The patient was treated with IV fluids and bowel decompression. Liver-spleen scan with 99m technetium-sulfide colloid revealed a large, lucent defect in the spleen (Fig. 1). Subsequent to the scan, approximately 16 hours after admission, the patient's physical signs changed. His heart rate increased to 168 beats/minute, his blood pressure increased to 140/80 mm Hg, and he developed marked generalized abdominal tenderness with diffuse guarding. His Hct fell to 29%. Peritoneal lavage revealed gross blood.

At exploratory laparotomy, through a supraumbilical midline incision, 600 cc of blood was removed from the peritoneal cavity. There was a diagonal laceration through the superior pole of the spleen, extending from the anterolateral aspect of the diaphragmatic border to the splenic hilum, 2 cm in depth (Fig. 2). The splenic tissue cephalad to the laceration was devitalized. All other intra-abdominal structures were free of trauma.

The splenic flexure, stomach and small bowel were packed away. The spleen was reflected medially, allowing for division of its lateral and inferior attachments. Division of the superior edge of the

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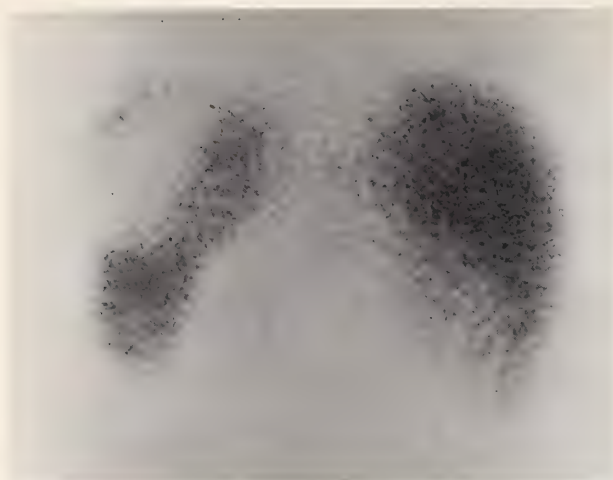


FIG. 1. Preoperative liver-spleen scan (posterior view) showing large subcapsular splenic hematoma.

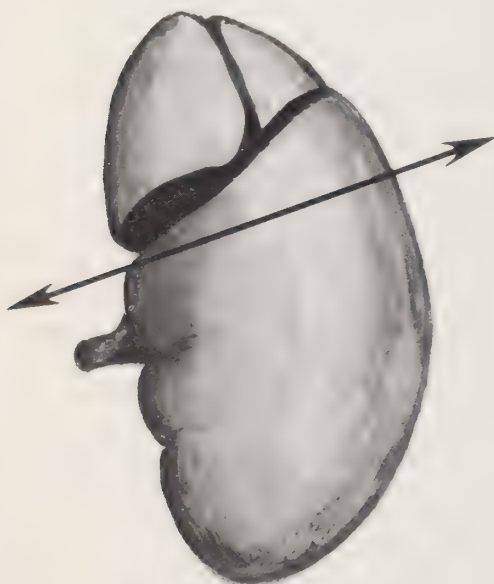


FIG. 2. Sketch of spleen showing laceration, arrow showing line of resection.

gastrolial ligament allowed for complete delivery of the spleen from the abdominal cavity. With the splenic vessels between the index and middle fingers of the left hand, the superior pole of the spleen was divided in a "fish-mouth" fashion using the bovie scalpel. Hemostasis of the raw splenic surface was then obtained by approximating the splenic capsule with through and through horizontal mattress sutures of 00 Chromic catgut (Fig. 3). Avitene[®] was used to achieve further hemostasis, and omentum

was secured over the repaired surface with 5-0 chromic sutures. This technique was comparable to those previously described by Burrington (13, 5) and Sherman et al (11).

The patient had an uneventful postoperative course. On the seventh postoperative day, a splenic technetium sulfur colloid scan demonstrated absence of the superior pole, but an otherwise normal splenic pulp (Fig. 4).



FIG. 3. Partial splenectomy with hemostatic sutures in place.



FIG. 4. Postoperative liver-spleen scan (posterior view) showing absence of superior pole.

Discussion

Currently, there are three modalities available for the management of splenic trauma. They are: splenectomy in all cases of splenic injury; nonoperative management with close observation and transfusions as needed; and splenorrhaphy. Recent trends in the literature have been away from splenectomy in all cases. This trend stems from awareness of increased susceptibility to and fatality from gram positive septicemia in postsplenectomized children. The real debate exists between the advocates of nonoperative management and those of splenorrhaphy. At issue are not cases of spleens damaged beyond repair, in which splenectomy is done out of necessity; but rather those patients who would qualify for splenic repair. It is this latter group of patients that are managed nonoperatively by the surgeons at The Hospital for Sick Children, Toronto.

In reviewing the studies from Toronto (1, 2, 14) one must take issue with a number of points. With the nonoperative management, some of the patients require prolonged hospitalization, with an average stay ranging from three to six weeks. On occasion, these children require intensive transfusion therapy with 50%–90% of blood volume being replaced. The surgeons are never able to completely evaluate the extent of injury in those patients treated nonoperatively. Of the total children observed, 35%–38% eventually undergo laparotomy. In support of the surgeon's clinical management at The Hospital for Sick Children, splenic function is preserved and surgery is avoided in most patients with splenic injury. In one of their series, 20 out of 22 patients show none to minimal residual defect of follow-up splenic scans (14). Finally, there are no reported cases of traumatic splenic pseudocysts or abdominal splenosis, and only two cases of delayed splenic rupture.

In comparison to the patients managed nonoperatively, the patients who underwent splenic repair had an average hospitalization of about one week, (6, 10, 11) and resumed full activity after one month.

The full extent of injury was accurately determined at the time of laparotomy, and no long-term complications have yet been reported.

Truly, the most successful management is the middle ground between nonoperative management and splenic repair. The surgeon must therefore decide at which point laparotomy is indicated. It is clear that all children with splenic trauma warrant initial observation and documentation of injury by liver-spleen scintiscan. Once the decision for laparotomy is made, the surgeon's approach should be towards preservation of splenic tissue.

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Seminars in Reproductive Medicine

Norbert Gleicher, M.D., Editor

Interaction of Ovarian Hormones with Normal Endometrium and Endometrial Adenocarcinoma

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Any discussion of the actions of steroid hormones on the endometrium requires the consideration of receptors since it is now accepted that they mediate the biologic action of these compounds. A description of interaction of estrogens with their receptors serves to illustrate principles that apply to other steroid hormones.

Evidence indicating that receptors were involved in estrogen action was provided in the late 1950's by experiments in which tritiated estradiol ($^3\text{H-E}_2$) of high specific activity was injected into immature rats and the radioactivity retained by different tissues was measured at various times after the injection (1). It was found that target tissues (uterus, vagina) retained the tracer much longer than non-target tissues and it was postulated that the presence of receptors was responsible for the retention of the hormone.

The concepts on mechanisms of action of the hormone prevailing by the end of the 1960's can be summarized as follows:

- Steroid receptors are tissue specific proteins which bind with high affinity, low capacity, and remarkable selectivity to compounds having similar biologic action.
- After binding the hormone, the steroid receptor present in the cytoplasm undergoes a temperature dependent transformation, detectable by

changes in sedimentation constants and chromatographic or electrophoretic behavior.

- The transformed or activated hormone-receptor complex translocates to the nucleus, binds to acceptor sites in the chromatin, and influences the process of transcription (2).

This scheme provided the rationale necessary to develop tests for the prediction of responsiveness of patients with breast cancer to surgical procedures aiming to eliminate estrogens from circulation (ovariectomy, adrenalectomy or hypophysectomy). Patients with breast tumors without estrogen receptors would not be expected to benefit from these major procedures, a prediction which was confirmed by clinical studies (3). The concept of involvement of receptors in estrogen action also led to the search for antiestrogens, that is, compounds which can compete for the binding of E_2 to the receptor but have not sustained estrogenic activity. These efforts were successful and one of these compounds, viz tamoxifen, is now widely used in the treatment of breast cancer patients with tumors containing estrogen receptors. It is difficult, however, to explain with this model the paradox that physiologic amounts of estrogen enhance tumor growth whereas pharmacologic doses cause remissions, or the fact that glucocorticoids, androgens and progestins are not less effective than estrogens as therapeutic agents.

Revisions of the simple scheme became necessary, as a result of extensive work carried out during the last decade.

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The postulate that steroid receptors are confined to the cytoplasm until binding the hormone and moving into the nucleus has been challenged by the demonstration of the presence of nuclear receptors not bound to the hormone in MCF-7 cells (4, 5), pig uterus (6), human myometrium (7) and in normal or neoplastic human endometrium (8). The main evidence for the existence of these hormone-free nuclear receptors is that they can be labeled by incubation with $^3\text{H-E}_2$ at 0–4° C for short periods of time, a condition under which there is no exchange between endogenous E_2 bound to the receptor and the exogenous labeled hormone.

Several forms of the estrogen receptor, besides the cytoplasmic and nuclear receptors originally described, are now being considered. Two types of high affinity, low capacity binders of E_2 were distinguished in rat uterus cytosol (and also in nuclear preparations) by resolving the curves of saturation with $^3\text{H-E}_2$ into two components (9). Heterogeneity in the cytosol receptors for E_2 in the rat uterus was also revealed by experiments in which it was shown that tamoxifen competes with $^3\text{H-E}_2$ binding to only one of two high affinity, low capacity binders (10). In none of these experiments was a distinction made between endometrial and myometrial components of the uterus and therefore the possibility that different receptors are present in different cell types remains to be explored.

Differences between cytosolic and microsomal E_2 receptor in pig uterus have also been described (11).

Heterogeneity in nuclear binding sites for estrogens were detected during studies on competition of estetrol and ethynyl estradiol for the labeling of human endometrium with $^3\text{H-E}_2$ (12). Furthermore, only a fraction of the estrogen receptor in nuclei is removed by extraction with 0.4 M KCl solutions and a distinction between salt-extractable and salt-resistant nuclear receptors can be made (9). Clark et al have proposed that the salt-resistant complexes might be tightly bound to sites in the chromatin where they exert their action on transcription, since biologic effects of the hormone correlate better with the concentration of these salt-resistant complexes than with the total concentration of nuclear estrogen receptors. They also proposed that the continuous presence of these receptor complexes for at least six hours is necessary to produce uterotrophic effects. Thus, estriol, which forms a receptor complex that is rapidly lost by the nucleus, does not produce uterine growth after a single injection but is active when injections are repeated at short intervals or when estriol-releasing pellets are implanted (9).

Another change in the outlook of receptors concerns their specificity towards different classes of active steroids. In contrast to previous beliefs, it has recently been found that in many instances some steroids, when their concentrations are sufficiently high, bind receptors that are primarily involved in the action of another group of hormones. For example, androgens can bind and activate estrogen receptors and progestins can interact with the glucocorticoid and androgen receptors.

Several problems related to the interaction of estrogen receptors with nuclear chromatin have been identified but remain unresolved. One of these is whether the nuclear sites binding receptor-estrogen complexes are tissue specific, are saturable and are homogeneous, and whether these sites are on nonhistone proteins, on DNA, or on both. Recent reviews describe experimental results concerning these problems (2, 13).

One of the serious gaps in the current understanding of hormonal action is the fate of the nuclear receptor complex. Horwitz and McGuire have proposed that estrogenic action may be related to "nuclear processing" of the receptor-estrogen complex. They describe that addition of E_2 to a culture of MCF-7 cells results in initial high levels of hormone-bound receptor in the nucleus but that the concentration declines in about five hours to a plateau 70% lower than the initial value, without any increase in the levels of cytoplasmic receptor or in the salt-resistant fraction of the nuclear receptor (14, 15). During this period, the hormonal effect is manifested in an increase in the levels of progesterone receptor. Nafoxidine, an antiestrogen devoid of estrogenic activity, can also increase initially the nuclear estrogen receptors to the same levels obtained with E_2 but they remain constant during the five-hour-period, not showing the decline to a lower plateau. Tamoxifen, a partial agonist which cannot elevate the progesterone receptor concentrations to the levels reached under E_2 stimulation, produces only a 50% decline of nuclear estradiol receptor levels during a five-hour period. Actinomycin D, an intercalating agent which inhibits RNA synthesis, does not affect binding and translocation of the receptor by E_2 but suppresses processing immediately after addition (16).

These results raise the question of differences between receptor-estrogen complexes formed by various estrogens. It is possible that these differences could be explained in terms of kinetics of dissociation of the hormone from the complex but structural differences in the complex itself should be considered. Another question related to the fate

of nuclear receptors is whether, and to what extent, they are returned to the cytoplasm and how the rate of synthesis of new receptors is regulated by the concentration (or processing) of the receptor in the nucleus.

Regulation of Estrogen and Progesterone Receptor Levels During the Menstrual Cycle

Levels of estradiol and progesterone receptors in human endometrium have been measured throughout the menstrual cycle.

The estrogen receptor is present at high concentrations (approx. 3 pmol/mg DNA) in proliferative endometrium and declines to one fifth of that level during the luteal phase (17, 18). In vivo experiments in which medroxyprogesterone acetate was administered for a few days to women in the follicular phase of their cycle showed that the physiologic fall in estrogen receptors is due to the action of progesterone secretion by the corpus luteum (19).

The progesterone receptor level is highest at the time of ovulation (18, 20). This pattern is understandable since estrogens, which are elevated during the follicular phase, have been shown to induce the progesterone receptor and progesterone, elevated during the luteal phase, impairs the action of estrogens and may even destroy its own receptor by a more direct mechanism (21).

Metabolic Regulation of Concentrations of Estradiol

Of importance for the analysis of the physiologic action of estrogens is the usually neglected consideration of the metabolism of E_2 in the target cell. Intracellular metabolism of the hormone can be as much a determinant of its action as is the amount of receptor present in the cell. Furthermore, both the enzymatic activity and the level of receptor can be subject to control mechanisms. Drastic differences in the rate of metabolism of E_2 are observed in endometrium during the follicular and secretory phases of the menstrual cycle. In superfusion experiments with ^3H - E_2 , the intracellular concentration of the labeled hormone at the steady state in secretory endometrium was about ten times lower than in proliferative tissue (22). In a series of in vivo and in vitro experiments, it was established that progesterone was responsible for these changes (23). The antiestrogenic effects of progesterone in human endometrium are two-fold: it lowers the levels of estrogen receptors and it increases the activity of the enzyme estradiol-17 β dehydrogenase, which catalyzes the conversion of E_2 to estrone (E_1),

a metabolite found not to be bound to the receptor in the nucleus in whole cell preparations (24).

Endometrial Cancer

Ovarian hormones have been related to endometrial cancer in several ways. The influence of estrogens on the development of endometrial cancer is revealed by the analysis of risk factors associated with the disease. Gusberg (25) reported evidence that led him to state that women exposed to prolonged estrogen stimulation in the absence of progesterone are at higher risk to develop endometrial adenomatous hyperplasia, a precursor of adenocarcinoma. Two conditions characterized by "unopposed" estrogenic environment were described in that article: the presence of estrogen-secreting ovarian tumors in postmenopausal patients and a history of prolonged administration of estrogens. It was subsequently indicated that other risk factors are also associated with the same hormonal pattern, viz. polycystic ovarian disease, infertility and obesity (26). In obesity, the conversion of endogenous androgens to estrogens is increased (27). The effectiveness of progestins in the treatment of endometrial cancer (28) further demonstrates that steroids can influence its development.

Application of the steroid receptor concept led Ehrlich and collaborators (29) to attempt correlating responsiveness to progestin treatment with the presence of progesterone receptors in the tumor. Using a cut-off point of 50 fmol/mg protein, they found in series of recurrent endometrial carcinomas, previously irradiated, a lack of response when progesterone receptor levels were below 50 fmol/mg protein and partial or complete response when the receptor levels were above the value.

Histologic grade is not a satisfactory predictive factor since a significant proportion of undifferentiated adenocarcinoma responds to progestin treatment and contains progesterone receptors (30).

Our approach to the identification of endometrial adenocarcinoma responsive to progestins is to administer medroxyprogesterone acetate for three to ten days, and to evaluate the responses histologically and biochemically. The biochemical parameters measured are the E_2 receptor levels and the activity of E_2 dehydrogenase. Changes were noted in many cases (31), indicating the feasibility of this approach. It remains to be determined, however, whether observed effects correlate with clinical response to steroid therapy. In a new series of similar experiments now in progress, progesterone receptors are measured in the specimens obtained before

treatment and their levels are related to the observed responses.

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Neurologic-Neurosurgical Notes

Allan E. Rubenstein, M.D. and Martin H. Savitz, M.D. Coeditors

Thalamic Infarction Producing Aphasia

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Abstract

A 62-year-old man with the sudden onset of a language disorder and mild right-sided motor and sensory deficit had a left anterior thalamic infarct on computerized scan. The patient's speech and language symptoms contrast with those previously described in association with extensive thalamic lesions, and more closely resemble features associated with aphasia secondary to cortical lesions. We suggest that the mechanisms proposed by others to account for the symptoms observed following thalamic hemorrhage may not be operative for smaller, more discrete lesions of the thalamus.

Aphasia is generally associated with a lesion in a quadrilateral or posterior frontal, superior temporal and inferior parietal cortex within the dominant hemisphere. Occasionally, patients with thalamic hemorrhage also have a language disorder (1-9). These patients are often described as inattentive, with numerous perseverations and paraphasic errors. We found no reported cases of thalamic infarction causing aphasia.

Computerized axial tomography (CT) has improved the recognition and localization of deep cerebral vascular accidents during life. Recently, we had the opportunity to treat and evaluate a patient with thalamic infarction demonstrated by computerized tomography. It is important to note that an infarction of the thalamus was associated with a language disorder and that our patient's language disorder was very different from the previously described entity of "thalamic aphasia," conforming, instead, more closely to the features of aphasia found with cortical lesions.

Case Report

A 62-year-old right-handed, bilingual hispanic male was admitted to the Neurology Service of the Bronx Veterans Administration Medical Center with a chief complaint of sudden onset of difficulty with speech and right sides weakness. The patient had a history of diabetes mellitus for the past six years treated with chlorpropamide. There was no prior history of hypertension or neurologic disorder.

On admission, blood pressure was 120/70 mm Hg. The general physical examination was unremarkable. Neurologic examination revealed an alert, oriented male with object-naming difficulty, slow spontaneous speech and paraphasic errors which were more evident in English than in Spanish. He was able to repeat well. No visual field defect was found with confrontation testing. The motor examination revealed flattening of the right nasolabial fold and 4+/5 weakness of the right extremities, more evident in the arm than the leg. Reflexes were symmetrical and both plantar responses were flexor. Sensory examination showed a slight decrement in sensation to pin on the right side of the body, including the face, as compared to the left side. Double simultaneous stimulation failed to reveal a consistent loss of sensation on the right side. All other sensory modalities were intact. There

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were no carotid bruits and the remainder of the examination was unremarkable.

Laboratory studies: Complete blood count, sedimentation rate and 6/12 channel chemistries were within normal limits except for an elevated glucose of 347 mg/dl. Cerebrospinal fluid: cell count, 5 RBC's, 4 WBC's (100% lymphocytes), Protein 42mg%, Glucose 145mg/dl. Serology of the CSF: nonreactive. CT scan of the brain revealed an area of lucency only in the anterior left thalamic region (Figs 1A, 1B). No other abnormalities were noted. Repeat CT scan approximately one month later revealed essentially the same findings (Figs. 2A, 2B). A technesium brain scan was normal.

Speech and Language Evaluation

An initial speech and language evaluation was given four days after admission. The patient was then re-evaluated at four and six weeks later. The dates of these evaluations coincide with the dates of the two CT scans. Results of both evaluations were virtually identical. Therefore, the general findings of both evaluations will be reported.

During these evaluations the patient was alert, oriented and cooperative. He engaged easily in con-

versation with little apparent difficulty in either auditory comprehension or verbal expression, with the exception of occasional phonemic paraphasic errors and mild word-finding difficulty. At times, however, he exhibited more significant impairment, such as being unable to give either his address or job title despite repeated attempts at doing so. A detailed examination did, in fact, uncover a significant language disorder.

Auditory comprehension of nonredundant verbal commands (Token Test) deteriorated with increasing length and complexity. Similarly, although oral reading and understanding of simple written questions showed no impairment, comprehension of lengthier and more complex written material was extremely poor.

He could produce prompt, well-formed utterances from target words on command, but did poorly at providing definitions. Performance on confrontation naming declined as a function of word frequency, as evidenced by an increase in response latency and associative responses. Repetition of words and sentences was intact.

The patient could write to dictation and produce grammatic self-formulated written sentences. How-

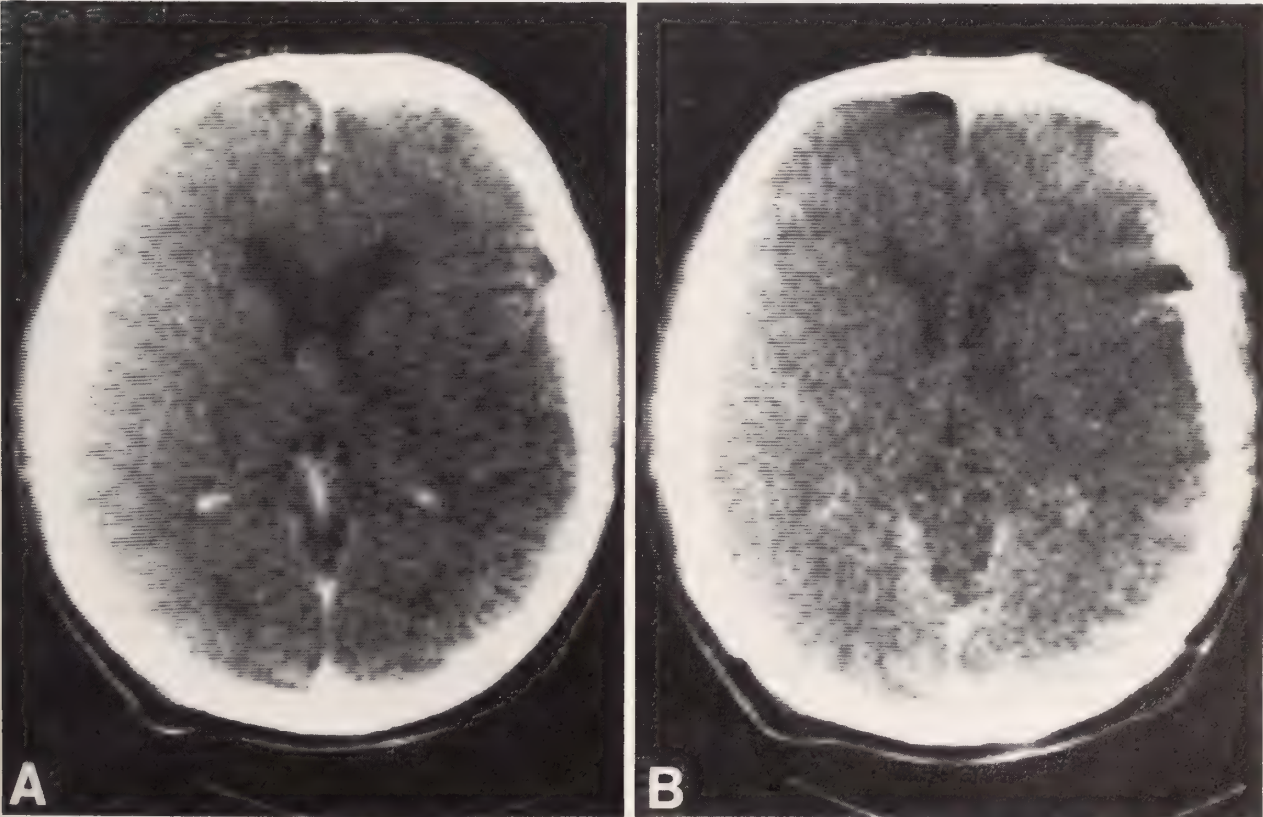


FIG. 1. CAT scan with contrast administration four days following admission. (A) (B) Views through the thalamic region reveal an area of lucency in the location of the left anterior thalamus and internal capsule.

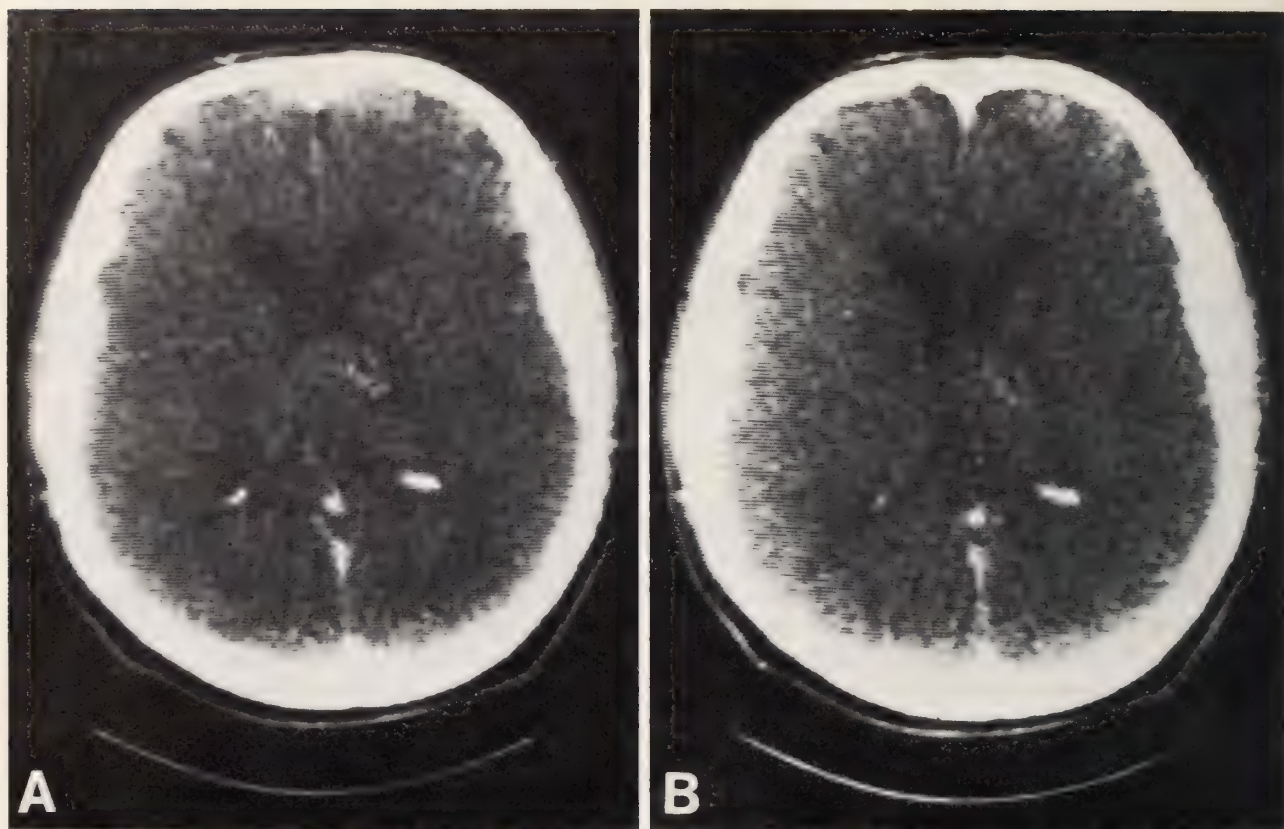


FIG. 2. CAT scan with contrast administration one month after the initial scan. (A) (B) Views as before; note same area of lucency with some enhancement. These scans were most consistent with an infarction.

ever, numerous spelling errors were noted both within sentences and for words in isolation (Fig. 3), which cannot be explained on the basis of bilingualism and educational level alone. Simple arithmetic operations were performed accurately and promptly.

The final impression was that of a mild to moderate aphasic disturbance crossing all language modalities, with increasing impairment evident as a function of linguistic demands.

Discussion

The exact role of the thalamus in language is an issue that has received wide attention, but for which the evidence remains equivocal. The pertinent question of recent years has not been whether damage to the thalamus of the dominant hemisphere may interfere with communicative function, but rather, what is the nature of this disturbance?

The majority of recent opinion is that thalamic lesions produce a communicative disorder qualitatively different from those observed secondary to cortical lesions (3, 6-8, 10). This conclusion is based primarily upon symptoms demonstrated by patients

with spontaneous thalamic hemorrhage (3, 6, 8, 9) and damage of the thalamus and its projections to the temporal lobe after surgery (7).

Ciemens (2) reported two cases of mixed aphasia following localized hemorrhage in the left thalamus. His first case, had a more thorough speech evaluation, demonstrated general somnolence with a tendency toward perservation, had difficulty with object naming, had decreased and fragmented spontaneous speech and comprehension and writing difficulties. Repetition was reportedly unimpaired.

Mohr, Watters and Duncan (3) reported the speech behavior of two patients with verified left thalamic hemorrhage and aphasia for whom two distinct states were apparent. During the first, these patients typically produced fluent speech characterized by jargon, logorrheic paraphasia, perseveration, echolalia and intact repetition. Vocal intensity was reported to wane as the patients became silent and somnolent unless prompted to continue to respond. In sharp contrast to this state was one in which these same patients were able to perform highly complex language tasks.

Reynolds et al (6, 8) reported five cases of left thalamic hemorrhage and aphasia. The character-

Porch Index of Communicative Ability

GRAPHIC TEST A

Name _____
Date 11/9/78
By CG

Speech Path. MF#299740
VA Med. Ctr. Bronx NY
Thalamus Study
Date of Photo: 8/22/79 md

13 With the fork, I can clean my mouth teeth.

12 I write a letter with my fountain pen.

11 The key is very important to me.

10 With the fork I can eat my food

9 Cigarette is a very bad habit.

Porch Index of Communicative Ability

GRAPHIC TEST B

Name _____
Date 11/9/78
By CG

Speech Path. MF#299740
VA Med. Ctr. Bronx NY
Thalamus Study
Date of Photo: 8/22/79 md

1 The name of the first article on the table is

10 a tooth brush.

9 pen

8 key

7 fork

6 cigarette

FIG. 3. Porch Index of Communicative Ability (modified), administered four days following admission. Patient's written responses for test items in full view (toothbrush, pen, key, fork, cigarette) regarding: (A) functions of objects, (B) names of objects, (C) names of objects after spoken by examiner, (D) names of objects after spelled by examiner.

istic speech symptoms included paucity of spontaneous speech with decreasing vocal intensity, anomia, perseverations and neologisms in the presence of intact comprehension and word repetition abilities.

More recently, Cappa and Vignolo (9) have reported three additional cases of aphasia following hemorrhage in the left thalamus in which clinical symptoms were characterized by a reduction of spontaneous speech, preserved repetition, and vary-

Porch Index of Communicative Ability

Speech Path. MF#29974A
VA Med. Ctr. Bronx NY
Thalamus Study
Date of Photo 8/22/79 md

GRAPHIC TEST C

Name _____
Date 11/9
By CRJ

tooth-brush
cigarette
pen
fork
key

Porch Index of Communicative Ability

GRAPHIC TEST D

Name _____
Date 11/9
By CRJ

toothbrush
cigar/cigarette
pen
fork
key

Fig. 3C-D.

ing degrees of semantic paraphasia, perseveration, anomia and auditory comprehension deficit.

The speech disturbance of Luria's surgical patient was characterized by "extraneous associations" and "inert stereotypes and perseverations," although no gross attentional disturbance could be demonstrated. The patient's responses were so far removed from either the phonemic or semantic targets that the term paraphasia seemed inappropriate.

The clinical picture of most of the reported cases of left thalamic hemorrhage and aphasia is strikingly similar. However, a number of different explanations have been offered regarding the possible role of the thalamus in language.

Penfield and Roberts (11) suggest that the role of the thalamus in the "elaboration of speech" is its "functional integration" and coordination of input to subcortical areas from the cortical speech areas.

Ojemann (10) attributes the thalamus with im-

portant alerting and attention focusing mechanisms, the destruction of which may disrupt mnemonic processes insofar as these mechanisms are important for input to and retrieval from verbal memory. In this way, then, he attempts to account for the observed fluctuations in performance, perseverations and neologistic verbal behavior of these patients, or what Reynolds (6, 8) calls the "hallmarks of 'thalamic' language disturbances."

Cappa and Vignolo (9), on the other hand, note a similarity between the speech characteristics of their patients and those of the so-called transcortical aphasias; that is, intact repetition, few, if any, phonemic errors in spontaneous speech, and occasional echolalia. They suggest that the role of the thalamus in language (like that of the marginal cortical speech zones) may be to "subserve the use of words as meaningful units" and thereby contribute to the "semantic level of verbal behavior," while adequate phonemic behavior may depend upon the integrity of other cortical or subcortical areas and/or mechanisms.

In support of Cappa and Vignolo, Mohr et al (3) maintain that "the intact repeating from dictation argues that the other features of the language performances might properly be considered wholly thalamic in origin." However, unlike the above authors, Mohr et al also feel that the high level, albeit brief, performance of their patients renders the notion of this as a linguistic disorder highly unlikely, viewing the logorrheic paraphasia more as a deregulation of cortical input than one of true linguistic impairment.

Luria (7), too, doubts the aphasic nature of speech symptoms secondary to thalamic involvement, suggesting that the disturbances of language following thalamic lesions do not reflect disturbances of linguistic structure, but, rather, represent defects of "vigilance" whereby the "specificity of the speech process was lost, the blocking of already evoked or extraneous associations became impossible and the selectivity of speech had broken down." In other words unlike the disturbance of verbal processes resulting from cortical lesions, he believes that these speech symptoms reflect a disturbance in gating and blocking processes.

Recalling Ojemann's (10) view of thalamic function in speech, it is clear that both he and Luria are describing a similar mechanism. Yet, for Ojemann the disorder is linguistic; for Luria the disorder is not. However, this is an issue that is not likely to be easily resolved, nor is it our intention to even address it here. Instead, we find it illuminating to

compare the speech characteristics of our patient with those of the other reported cases, since he differs from these patients in a number of significant ways.

First, almost all other reported cases demonstrated a type of inertia or lack of communicative drive, often evolving into unresponsiveness in the more severe cases, but almost always requiring prompting by the examiner. The paucity of spontaneous speech is more likely the by-product of this apparent lack of motivation or decreased arousal than it is likely to be a reflection of general fluency. In sharp contrast to this, our patient's verbal output was neither sparse nor prompted, except possibly in instances of extreme frustration.

Second, paraphasia and perseveration by no means typified the speech of our patient, whereas these appear to be the common denominators or "hallmarks" of the speech of the other reported cases. In other words, our patient appears to exhibit a language disturbance that is qualitatively different from that reported following hemorrhage of the thalamus of the dominant hemisphere.

Therefore, while certain speech symptoms may be the common threads of language disorders following thalamic hemorrhage, considerable caution must be exercised in attempting to define the function of the thalamus from such cases. One must bear in mind that damage following hemorrhage is often of rather large proportions, as in one case reported by Ciemens (2), where a 2.5 centimeter lesion included an area extending from the anterior nuclear group to the posterior commissure; in other words, the entire thalamus of that hemisphere. Since it is likely that the thalamus is a functionally heterogeneous structure given the numerous intrinsic and extrinsic connections of its nuclei, it would be an ambitious venture to either assign or deny it a specific role in language when damage is so extensive and adjacent effects of compression and edema are unknown. Clearly, lesions of this type do not lend themselves to establishing correlations between morphology and function. Therefore, while it is interesting to note the manifest speech behaviors secondary to such lesions, the role of the thalamus in language can hardly be inferred.

The systematic assessment of speech symptoms following smaller, more discrete, spontaneous lesions of the thalamus of the dominant hemisphere, such as infarction, may provide more information regarding the role of the thalamus in linguistic processes. Our case is a right-handed man with an infarction of the anterior thalamus of the left hem-

isphere who does not have a disturbance of speech previously considered to be characteristic of thalamic lesions, but rather has a mild to moderate aphasia resembling those produced by cortical lesions. To our knowledge, this is the first such case to be reported. It is not our intention, however, to localize a particular language function in the thalamus, or to even begin to propose a mechanism by which such a language disturbance might be produced. Rather, this case demonstrates that the speech symptoms previously considered to typify "thalamic language" may, in fact, not be representative of thalamic involvement, except possibly when destruction of the thalamus is nearly complete.

The routine use of CT scan in the evaluation of the stroke patient may uncover more patients with thalamic infarction and an aphasia similar to that of our patient which, in the past, would have remained unrecognized. It is, therefore, imperative for the clinician to carefully evaluate the stroke patient's speech performance and to compare this with the anatomic localization afforded by computerized tomography. The recognition of thalamic infarction during life and the thorough clinical evaluation of these patients may then help provide us with further insights and answers regarding the role of the thalamus in language.

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Ophthalmologic Notes

Keith M. Zinn, M.D., Editor

Peripapillary Subretinal Neovascularization in Presumed Ocular Histoplasmosis: A Case Report

PAUL R. ROSENBERG, M.D.* AND KEITH M. ZINN, M.D.**

Presumed ocular histoplasmosis (POH) is a clinical entity classically characterized by the following triad (1-7): 1) peripapillary choroiditis, 2) peripheral atrophic chorioretinal scars (spots) and 3) subretinal neovascularization. Some authors (8) have added the presence of a positive histoplasmin skin test (positive in approximately 90% (9)) to this diagnostic criteria, while most authors have been content with less than all three of the classic fundus findings (10).

We have recently seen a young black male who had peripapillary hemorrhagic choroiditis, subretinal neovascularization, and a significantly elevated serum histoplasma capsulatum complement fixation antibody titer.

Case Report

D.H. is a 37-year-old negro male who had blurred vision in the left eye of at least four weeks duration unaccompanied by any headache, ocular pain, fever, or general malaise. Born in New York, the patient had never traveled outside the greater New York area during his lifetime. The patient was initially seen by a neurologist who found a central scotoma in the left visual field and an otherwise completely

normal neurologic examination. An ophthalmic consultation and examination revealed the following:

The best corrected visual acuity was 20/20 in the right eye and 20/400 in the left eye. Manifest correction could not improve the visual acuity in either eye. Extraocular motility studies revealed full rotations of either globe in all standard fields of gaze and the eyes were straight in the primary position of gaze. Cranial nerve function II, III, IV, V (ophthalmic division), VI and VII were intact bilaterally except for a demonstrable central scotoma in the left eye on Goldmann visual field testing to the I/3 white test object (Fig. 1). Amsler grid testing was positive for metamorphopsia in the left eye.

The lids, ocular adnexae and lacrimal gland apparatus as well as the conjunctivae, corneae and sclerae were within normal limits O.U. The intraocular pressures were 17 mm Hg in the right eye and 18 mm Hg in the left eye by applanation tonometry. Slit lamp examination of the anterior chambers revealed them to be clear, of moderate depth with grade III-IV anterior chamber angles for 360° O.U. The irides were brown and the pupils were equal, round and reactive to light and accommodation. The lenses and vitreous cavities were clear and unremarkable.

Funduscopy examination revealed blurred optic nervehead margins and the cup/disc ratio, in the horizontal meridian was less than 0.1 O.U. The left optic nervehead appeared slightly more elevated than the right optic nervehead. No spontaneous

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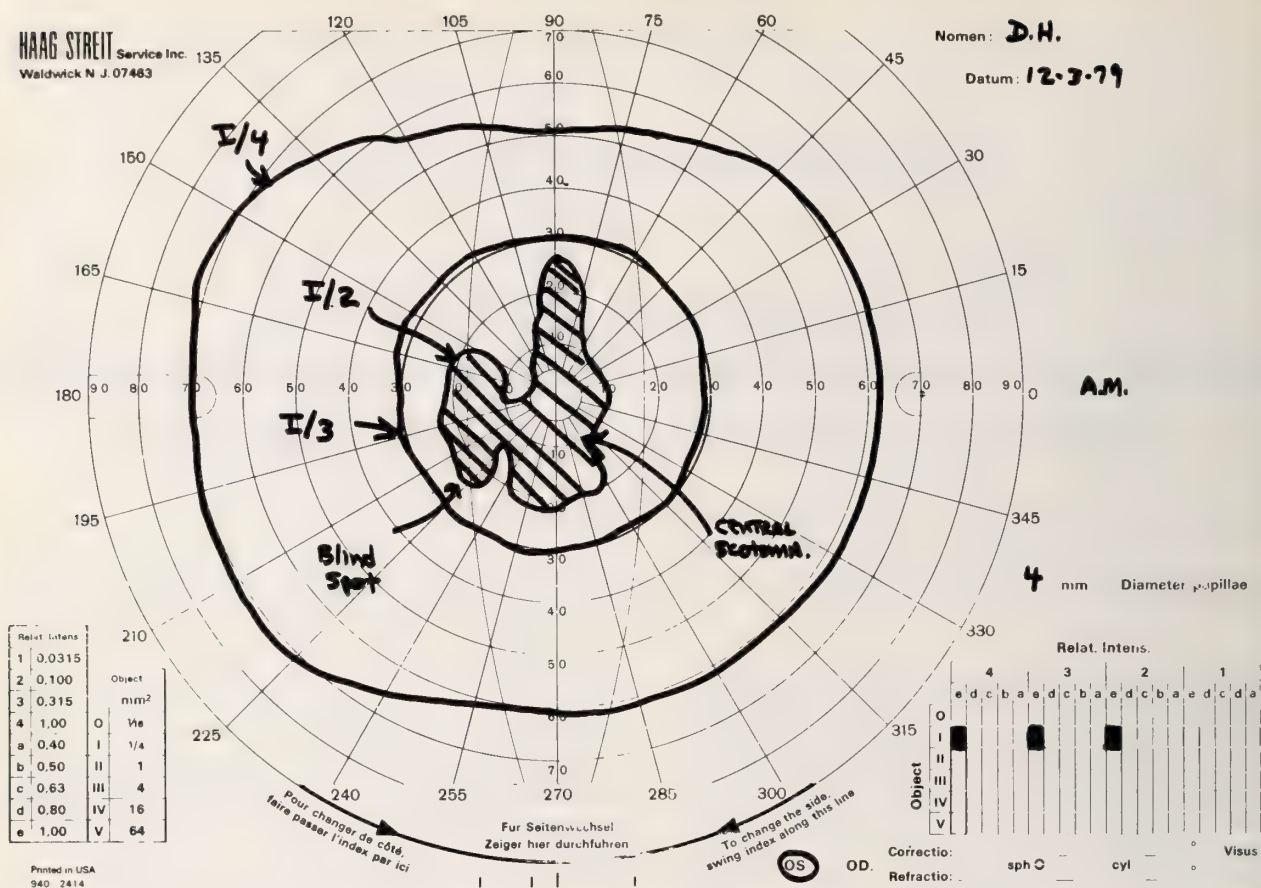


FIG. 1. Goldmann visual field showing a dense central-ecal scotoma (shaded area) to the I/3 white test isopter in the left eye of the patient.

venous pulsations of the disc vessels were noted O.U. However, light digital pressure applied to the wall of the globe did elicit venous pulsations of the disc vessels O.U. The macula in the right eye was within normal limits. The left macula revealed a greyish subretinal membrane with some subretinal hemorrhage at its edges (Fig. 2). The subretinal membrane extends from the superotemporal border of the optic nervehead to the upper macular region, ending in the perifoveal zone O.S. The vein/artery ratio near the optic nervehead border was 3:2 O.U. Peripapillary choroidal atrophy was noted O.U.

A and B scan ultrasonographic studies were within normal limits in both eyes. A rapid sequence fluorescein angiographic study revealed prompt retinal arteriolar filling with early hyperfluorescence of the peripapillary subretinal membrane which increased in its fluorescence in the later stages of the angiogram (Fig. 3). The subretinal hemorrhage remained as a hypofluorescent lesion blocking underlying choroidal fluorescence throughout the angiogram (Fig. 3). These fluorescein angiographic findings were consistent with a subretinal membrane and subretinal hemorrhage in the left eye.

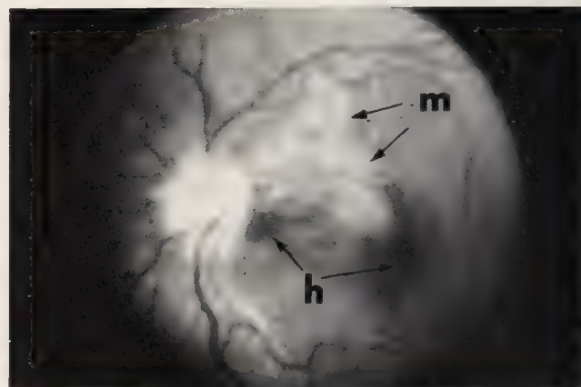


FIG. 2. Fundus photograph of patient's left disc and macula with a subretinal membrane (m) and some subretinal hemorrhage (h). The subretinal membrane begins at the temporal edge of the optic nervehead and extends temporally to the foveal region. Peripapillary choroidal atrophy is also present.

Peripapillary choroidal atrophy was also present on fluorescein angiography in both eyes. The fluorescein angiogram on the right eye was otherwise within normal limits.

Additional laboratory studies that were negative included the following: CBC, VDRL, ESR, LE

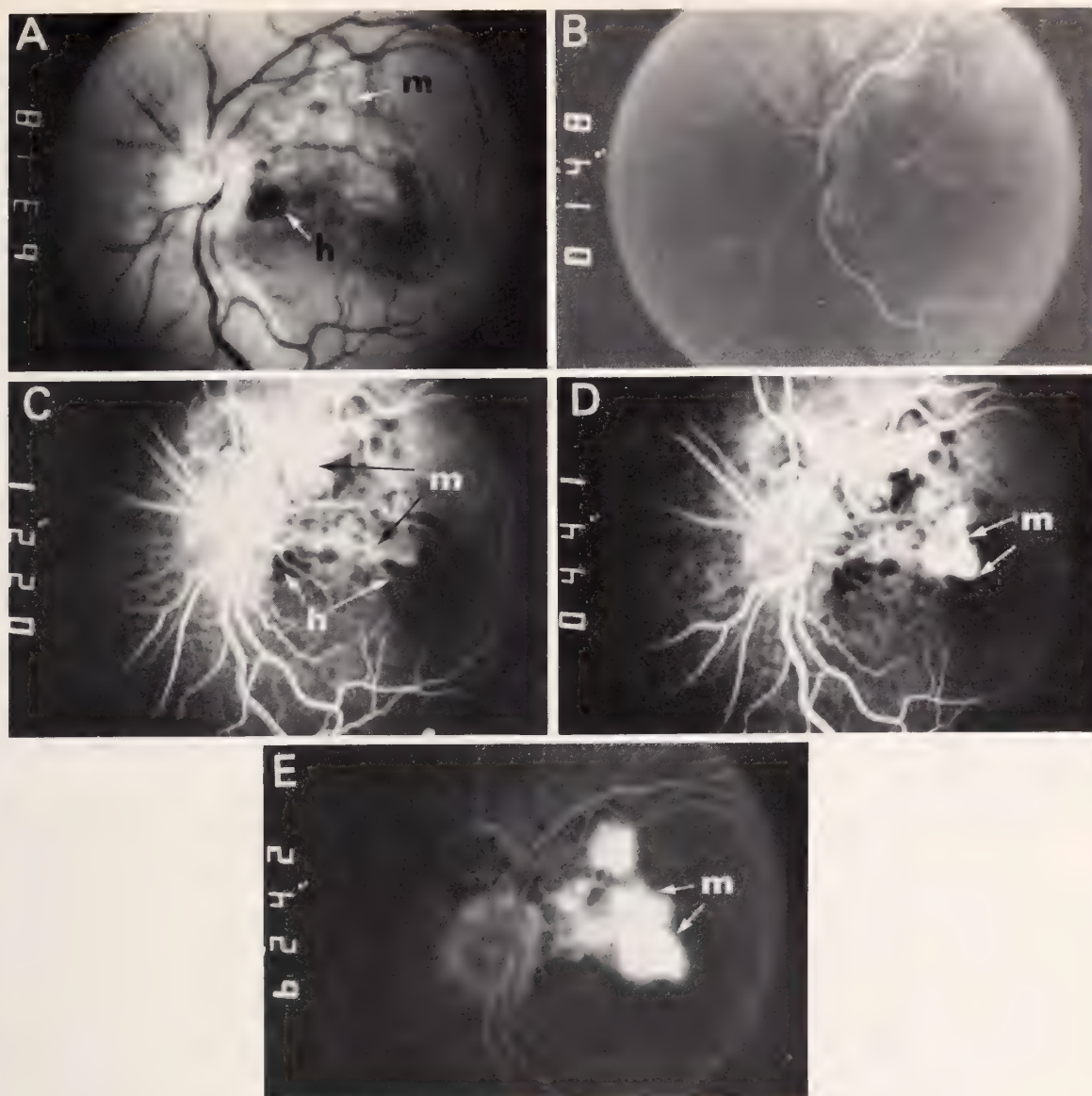


FIG. 3. Fluorescein angiogram left eye: A. Red free fundus photo left eye note greyish subretinal membrane (m) and subretinal hemorrhage (h) between the temporal border of the optic nervehead and the fovea. B. Early retinal arterial filling phase. C. Early retinal arteriovenous filling phase in which the subretinal membrane is hyperfluorescent (m) and the subretinal hemorrhage (h) is hypofluorescent blocking the underlying choroidal fluorescence. D. Mid-retinal arteriovenous phase note the hyperfluorescence of subretinal neovascular membrane (m) at foveal zone. E. Late retinal venous phase with staining of subretinal neovascular membrane (m).

PREP, rheumatoid factor, antinuclear antibodies (ANA), serum titers for toxoplasmosis, blastomycosis, cryptococcosis, coccidiomycosis, spinal tap with CSF cultures, and blood cultures. Skull x-ray, EEG and CAT scan were negative as well as a conjunctival biopsy. A PPD 5 and 250 strengths were negative.

However, a histoplasma complement fixation test for yeast phase antigen was positive in a titer of 1:32. HLA typing was not performed.

The patient was started on systemic steroid therapy consisting of 4 mg Decadron P.O. Q 6 hours for

a ten-day period and the visual acuity in the left eye improved from 20/400 to the 20/200 level. There was no further improvement in the vision and the patient was gradually tapered off the systemic steroids over the next several weeks.

Discussion

Presumed ocular histoplasmosis is an uncommon disease in negroes. Schlaegel (11) found that only 0.7% of patients with POH, in his series, were negro. In addition, the presence of POH in an area such as New York is considered to be uncommon (12). It is

also interesting to note the macular involvement in our patient, considering that in patients with POH there is only a 1:6 ratio of macular involvement in blacks vs whites, respectively.

Patients with disciform macular scars with the POH syndrome have shown a high frequency of HLA-B7 antigen (11, 13-15). In contradistinction, there was no significant increase in the frequency of this antigen in POH patients with only peripheral atrophic scars and without any disciform disease (14). Recently, HLA-DR w2 antigens have been found to be associated with the POH syndrome both in patients with peripheral chorioretinal atrophic spots as well as those with disciform macular scarring (15).

Schlaegel (1) describes four types of optic nerve-head changes in POH. These are: 1) diffuse, 2) nodular, 3) mixed, and 4) hemorrhagic. The hemorrhagic type is seen in only 15% of those cases with peripapillary choroiditis and is the only category in which symptoms are present. There is usually a demonstrable central scotoma, metamorphopsia and an absence of an arcuate scotoma. The case presented in this paper appears to be consistent with Schlaegel's description of a hemorrhagic peripapillary choroiditis. Of the nine cases reported by Schlaegel (1), five recovered useful central vision, and four were left with visual acuity of 20/200 or less.

Meredith and Aaberg (17), published a series of five patients (eight eyes) with peripapillary histoplasmic choroiditis in which there was subretinal neovascularization and hemorrhage in the peripapillary region similar to our case. Their fluorescein angiographic findings (17) were similar to ours and that of Hyvärinen, Lerer and Knox (18) and Gass (19). Argon laser photocoagulation had been used (17) in several cases with limited success. Many of the patients are prone to further subretinal hemorrhage and the formation of an organized disciform scar with visual deterioration regardless of treatment.

In the presumed ocular histoplasmosis syndrome, the evidence of *Histoplasma capsulatum* as the etiologic agent is based on clinical (3) and epidemiologic evidence (20-23). The reason for this is because the organism, to date, has not been identified histopathologically in the eyes of patients who fulfill the clinical criteria of the POH syndrome.

Our patient had a significant *Histoplasma capsulatum* yeast phase antigen titer of 1:32. Two weeks later his titer had dropped to 1:16. The *Histoplasma capsulatum* complement fixation test, when positive in titers of 1:8 or greater is presump-

tive evidence of infection (24). The higher the titer, the greater the likelihood of severe disease. Buechner et al (25) stated that titers of greater than 1:16 offer strong presumptive evidence of an infection with histoplasmosis. A note of caution concerning the complement fixation test stems from the cross reaction with other fungal diseases, such as blastomycosis and coccidiomycosis. However, these diseases are not associated with subretinal neovascularization (22), and specific testing of our patient's serum for these other fungi was negative.

In summary, patients with the POH syndrome who present with peripapillary subretinal neovascularization can go on to subretinal hemorrhage and disciform macular scarring and are difficult problems in terms of therapeutic management. The argon laser photocoagulation approach has met with limited success in this disorder.

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Seminars in Physiology

Walter Scott, M.D. and Melanic H. Cobb, Ph.D., Coeditors

Integration and Its Relation to Ocular Compensatory Movements

THEODORE RAPHAN, PH.D. AND BERNARD COHEN, M.D.

The oculomotor system uses both visual and vestibular information to maintain stable images on the retina. The vestibular system contributes to the stabilization by inducing eye movements that compensate for head movement. This occurs over the vestibulo-ocular reflex arc (VOR). The visual system generates slow eye movements that can either complement or suppress the VOR. Together, they maintain the angle of gaze when either the individual or the visual surround is moving. Recent research has demonstrated that the vestibular system performs several mathematical integrations that help produce an internal signal in the central nervous system that represents the relative velocity of movement between the head and the external visual surround. These integrations are necessary if there is to be adequate ocular compensation over a wide range of head movements. The purpose of this paper is to show how these integrations are accomplished and what role they play in contributing to ocular stabilization. By modelling these processes, it has been possible to gain insight into the organization of the VOR that is consistent with experimental data obtained from single unit recordings.

Signal Processing in the Semicircular Canals

Steinhausen (1, 2) described how the semicircular canals behave as accelerometers in responding to

angular movements of the head. However, Steinhausen's equations predict that the mechanical characteristics of the canals (the endolymph and the elastic properties of the cupula) convert head acceleration into a signal that is related to head velocity over the normal range of head movement. That is, the canals perform an integration by converting head acceleration to head velocity.

Recordings from fibers in the eighth nerve indicate that there is an integration of angular acceleration of the head by the semicircular canals. Afferent vestibular neurons have a resting discharge of about 50-100 spikes/second. In response to head rotation, the frequency of firing is increased or decreased (3-6). If the stimulus is a step of angular acceleration (a steady increase in head velocity), units in the vestibular nerve are modulated as shown in Figure 1A (7). Initially, firing rate increases, but as acceleration continues, the increase in firing rate becomes progressively smaller until it comes to a steady state level. At the end of acceleration (when head velocity is maintained at a constant level), firing rates fall to the resting level over a time course that is approximately exponential (Fig. 1B)(8). Thus, even though the head is still moving, the vestibular end organ signals that it has come to rest.

This process can be modelled mathematically by the response of a non-ideal integrator (a "leaky" integrator) to a step in angular acceleration (Fig. 1C, 1D). In the model, r represents the magnitude of the acceleration and $1/h$ is the time constant or the "leak" associated with the integrative processing. The interpretation of semicircular canal function in terms of this simple model is as follows: at the initiation of the stimulus the rise in activity is

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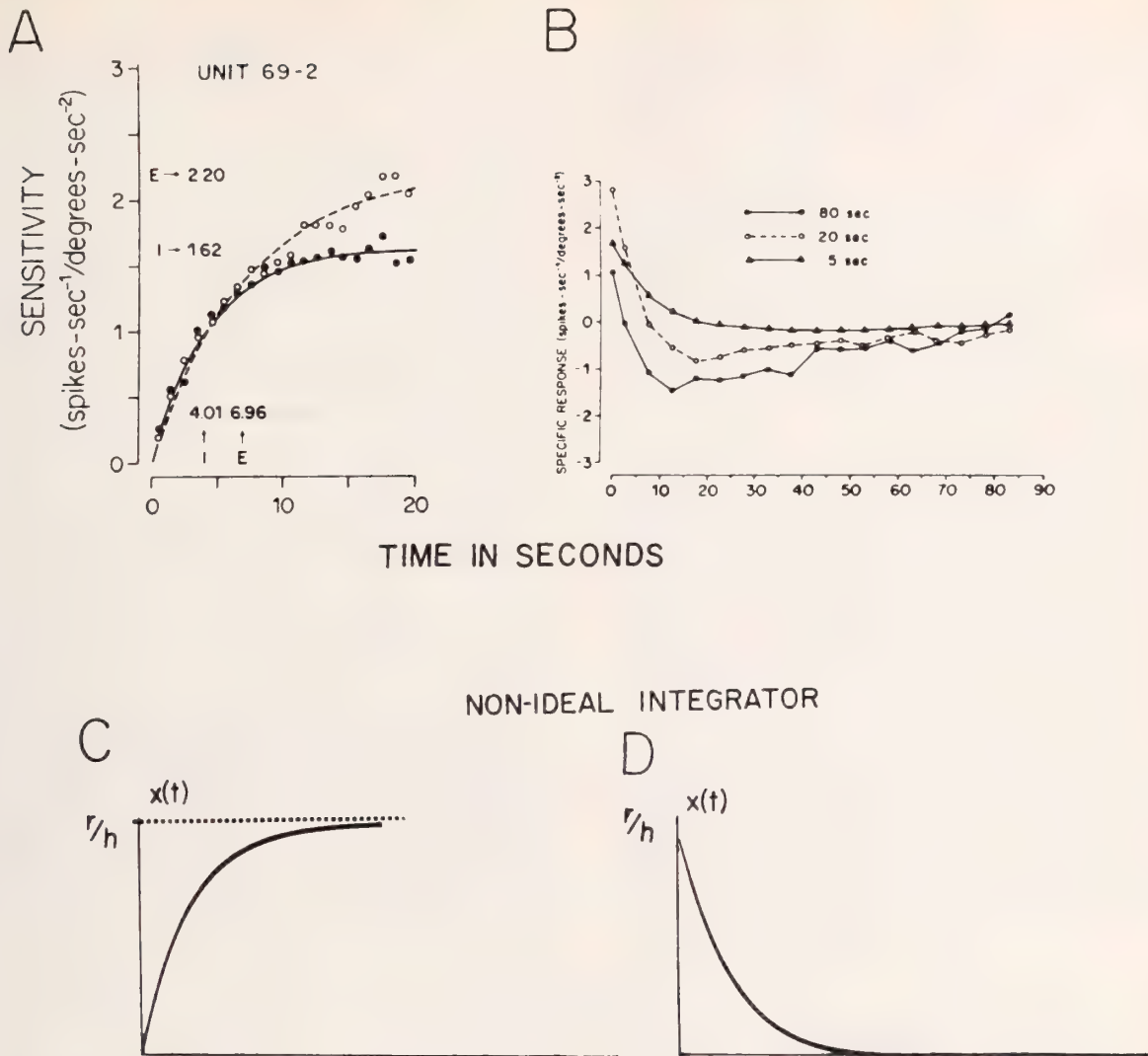


FIG. 1. A. Time course of the excitatory response (open circles) and inhibitory response (solid circles) of a horizontal canal unit. Curves were fit with exponential approximations. The vertical arrows show the sensitivity factors of the excitatory, and inhibitory responses. B. Time course of recovery of a superior canal unit from excitatory accelerations. Each curve represents the response following an acceleration of specified duration. Accelerations lasted for 80 sec at 3.75 deg/sec/sec; 20 sec at 15 deg/sec/sec; and 5 sec at 60 deg/sec/sec. C, D. Response of a non-ideal integrator to the onset (c) and end (d) of a step of excitation.

close to being linear (Fig. 1C). In this range, the semicircular canals behave as perfect integrators. However, during continued acceleration, shown by the dotted line in Fig. 1C, the leak becomes prominent, and the activity levels off to a steady frequency despite the continued increase in head velocity. At the conclusion of stimulation (Fig. 1D), acceleration goes to zero and head velocity is maintained at a constant level. The leak associated with the integrator causes a continuous decline in unit activity back to its resting level, (Fig. 1B, 1D). This is different from the response of an ideal integrator which would maintain or store the activity indefinitely.

Processing in the Central Vestibular System

Figure 1D implies that during head movements that accelerate rapidly to a constant velocity, activity in the vestibular nerve would rise rapidly and produce adequate compensatory eye movements over the first few seconds. This would be enough to compensate for most normal head movements. However, if head movement continued for longer periods, eighth nerve activity responsible for ocular compensation would fall and, after 10–12 seconds, the semicircular canals would signal to the brain that the head was stationary, although it was still in motion. Despite the inability of the peripheral receptor to hold a steady signal during continued

head movement, nystagmus, the ocular compensatory response to such movement, persists for much longer than several seconds. Typical nystagmus of the alert monkey induced by rotation at constant velocity in darkness is shown in Figure 2A. The top trace is the electro-oculogram (EOG) showing eye movement. The second trace is a differentiation of the EOG showing the velocity of the slow phases of nystagmus. (Slow phase eye velocity is a common measure of the "gain" of the VOR. The gain is equal to eye velocity/head velocity. If eye velocity equals head velocity, then the gain is one and ocular compensation is perfect.)

The time course of the decline in activity in the oculomotor system after a step in head velocity is shown by the envelope of slow phase velocity (SP Vel) (Fig. 2A). The induced nystagmus lasts for 30–60 seconds, with the velocity declining steadily during this time. This can be contrasted to the time course of the decline in activity in the vestibular nerve to a similar stimulus (eighth nerve time constant) (Fig. 2A). Eye velocity would follow this course if it were being driven only by the semicircular canals. The prolongation of the ocular response beyond the time when there is activity in the vestibular nerve indicates that further processing had taken place in the vestibulo-ocular reflex to support compensatory eye movements (10). That

is, the dominant time constant of the velocity command signal had been lengthened centrally. The functional consequence of this would be to promote better ocular compensation in darkness at low frequencies of head movement.

Modelling of Vestibular Nystagmus

Substantial advances in understanding of oculomotor function have come from modelling the oculomotor system based on control system theory. Models can quantitatively demonstrate the feasibility of certain postulates about the signal processing involved in producing nystagmus and other types of eye movements (See Raphan & Cohen, 1978 for a recent review of this subject). To model vestibular nystagmus two important aspects of the response must be considered: There must be a rapid response time after a sudden change in head velocity since compensatory eye velocities are reached in 5–25 msec. There must be a slow rate of decay of eye velocity during continued rotation in darkness once the eyes have reached their maximum velocity.

The simplest way to incorporate these characteristics is to have the input signal, the neural activity coming in over the eighth nerve, go both to an integrator and to a direct pathway that extends around it. The eye velocity command, that is the signal that drives the oculomotor system, would be

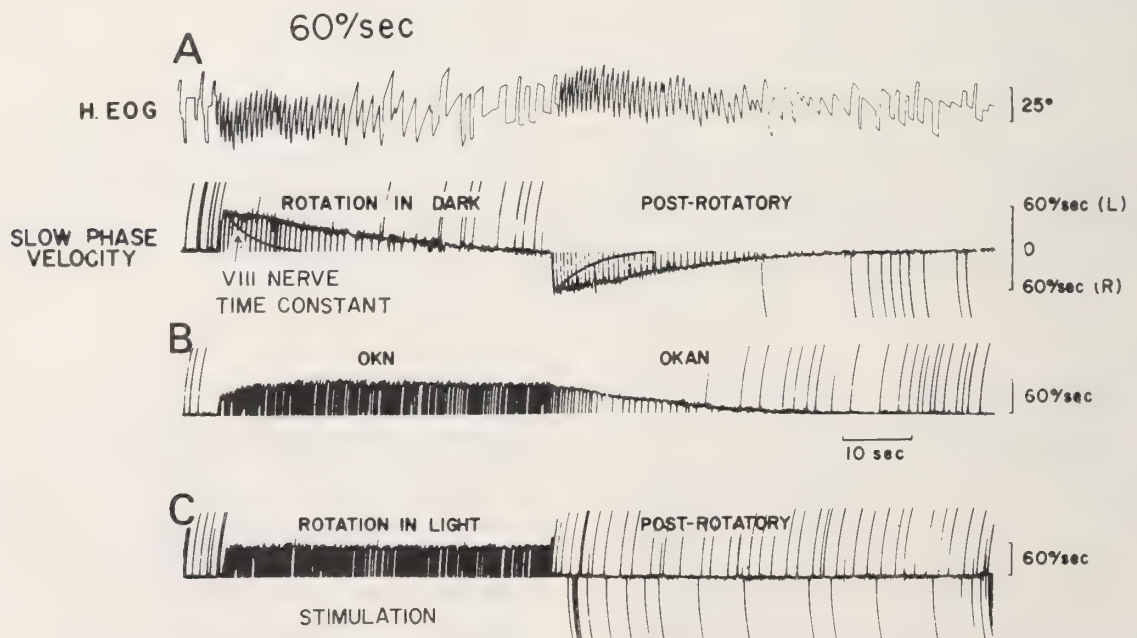


FIG. 2. A. Nystagmus induced by a step of platform rotation in dark (a), by a step of surround rotation (b), and by a step of platform rotation in light (c). The top trace in a is the horizontal EOG. The other traces are slow phases velocity. The stimulus velocity was 60 deg/sec/sec in each instance. Note that when rotatory nystagmus and OKN were in the same direction, their afterresponses were oppositely directed (a and b). Also note there was only a slight post-rotatory response to the right after rotation in light (c). The dotted line in the lower trace in A (VIII nerve time constant) shows the time course of eye velocity, that would be expected from a response to a step in velocity with only the VIII nerve driving the oculomotor system.

a sum of these two signals. Initially, activity in the eighth nerve reaching the eyes via the direct pathway would be the major source of the velocity command signal, and would be responsible for the short response time of the VOR. As the eighth nerve activity also excites the integrator, activity would be stored that would continue to drive the oculomotor system after the eighth nerve signal had fallen to zero. The slow falling time constant of the integrator would be responsible for the slow rate of decay during rotation. Since the function of this integrator is to store velocity information and its output contributes to the command signal for eye velocity, we have referred to it as a velocity storage integrator. This is to distinguish it from another neural integrator in the oculomotor system that converts eye velocity signals to eye position signals for use by the motoneurons (11).

Central Integration in Visual-oculomotor and Visual Vestibular Reactions

It has become apparent from studying the velocity characteristics of optokinetic nystagmus (OKN) and optokinetic after-nystagmus (OKAN) that the velocity storage integrator is also utilized in visual-oculomotor responses (12). Optokinetic nystagmus is induced by movement of the visual surround. (OKN can also occur when watching objects moving in a part of the visual field but the most effective stimulus for OKN is movement of the entire visual surround or full-field movement (13)). As full-field movement occurs primarily during head movement, OKN can be considered as the visually-induced counterpart to vestibular nystagmus induced by head movement. The two responses complement each other. For example, for head movement to the left vestibular nystagmus is induced with quick phases to the left, but the visual field moves to the right on the retina, inducing leftward OKN. By a combination of the two responses, the gain of ocular compensation is raised, stabilizing images on the retina.

During OKN (Fig. 2B), there is a rise in eye velocity to a steady state level that is close to the velocity of the visual surround. This velocity is maintained for as long as the stimulus persists. At the end of stimulation, if subjects are in darkness, there is persistent after-nystagmus, optokinetic after-nystagmus (OKAN), that lasts for up to a minute or more (Fig. 2B). The persistence of nystagmus indicates that the nervous system had stored activity related to slow phase eye velocity during OKN that was subsequently used to drive the eyes during OKAN. As can be seen in Figures

2A and 2B, the time course of the decline in slow phase velocity is similar for vestibular and optokinetic after nystagmus. This suggests that the same integrator is used by both the visual and vestibular systems (14).

There is other evidence to suggest that this is the case. The postrotatory response to rotation at a constant velocity is weaker if subjects are rotated in light, rather than in darkness (Compare Figs. 2A & 2C) though the cupula must have been deflected similarly at the end of rotation. Ter Braak (15) noted that the after-nystagmus for vestibular or optokinetic stimuli that induced nystagmus in the same direction, were oppositely directed. Thus, for vestibular and optokinetic stimuli that cause nystagmus to the left, OKAN is directed toward the left and postrotatory nystagmus is directed to the right (Fig. 2). He suggested that OKAN had summed with the postrotatory vestibular response to cancel it. A considerable amount of evidence indicates that this cancellation utilizes the velocity storage mechanism (14).

Model of VOR-OKN Generation and of Visual-vestibular Interaction

The preceding studies have led to the postulate that the velocity storage integrator superposes velocity signals from the visual system with those from the peripheral vestibular apparatus and outputs a velocity command. They also indicate that the same central integrator is used to store activity related to eye velocity during vestibular nystagmus, OKN and OKAN (Raphan et al, 1979). In support of this idea, a model that utilizes a common integrator and direct pathways from the visual and vestibular system predicts slow phase velocities of vestibular nystagmus, OKN, OKAN, and the visual-vestibular interaction.

In the model, vestibular nystagmus and OKN are produced by combined activation of direct and indirect pathways. The indirect pathways includes a non-ideal integrator, the velocity storage mechanism. Activity in the direct and indirect pathways summates to form the eye velocity signal that drives the eyes (14). Model simulation of the data shown in Fig. 2A-2C is presented in Fig. 3. Figure 3A shows the eye velocity predicted by the model in response to a step of vestibular excitation in dark. The overall characteristics are similar to those of the envelope of slow phase velocity of the vestibular nystagmus (Fig. 2A). Figure 3B shows the activity that would be predicted in the eighth nerve (cupula) and velocity storage integrator if they were excited

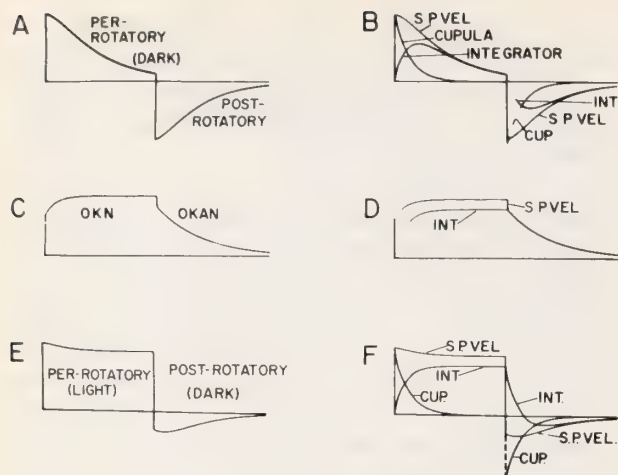


FIG. 3. Model predictions of slow phase eye velocity for a step of angular velocity in darkness (A), for a step of surround velocity (C) and for a step of angular velocity in light (E). B, D, F, Comparative changes in slow phase velocity, cupula deflection, and output of the velocity storage integrator for the responses shown in A, C and E, respectively.

by a step of head velocity. This activity is combined to give slow phase eye velocity (SP Vel). Because the cupula signal is falling while the integrator activity is rising, there is a short period when the velocity of the ocular response is approximately constant. This short plateau in eye velocity at the onset of vestibular nystagmus was recently shown to be present both in monkeys (Fig. 2A)(14) and humans (16).

Consistent with the idea that the same integrator is utilized for OKN and OKAN as for vestibular nystagmus, the model also simulates OKN and OKAN (Fig. 3C, 3D). As in the experimental data (Fig. 2B), there is a rapid and slow rise in eye velocity during stimulation and a slight fall in velocity at the onset of OKAN (12). Figure 3D shows a comparison of the integrator response and the overall response. The difference between them is due to the contribution made by direct pathways from the visual system.

The response of the model to a step in head velocity in light, that is, to rotation in a stationary lighted environment, is shown in Fig. 3E, 3F. The direct vestibular pathway is activated and immediately drives the eyes to the velocity of rotation. Eye velocity is maintained by the optokinetic response for as long as rotation continues. Because of the summation of stored activity in the integrator with activity coming in over the eighth nerve, there is only a weak postrotatory response when stimulation ceases (Compare Figs. 2A, 2C & 3A, 3E). It should be noted that this is the appropriate compensatory response. That is, the eyes move imme-

diately at the velocity of rotation, hold this velocity while movement continues and stop moving when rotation ceases.

Figure 3F shows the contribution of the integrator (Int), the eighth nerve (Cup) and the direct pathways to the total model response (SP Vel). The model predicts that after-nystagmus following rotation in light in a stationary surround would always be in the postrotatory direction. This is because the response is dependent on a summation of activity in the direct vestibular pathways with that in the integrator. The integrator can theoretically only be charged to 0.85 of stimulus velocity while the direct pathway has a gain close to one.

The Model and Its Relation to Unit Activity in the Vestibular Nuclei

It is not entirely clear how the vestibular nuclei participate in generation slow phases of nystagmus. It is clear, however, that there is a close link between the velocity storage integrator and unit activity in the vestibular nuclei. In the alert animal vestibular nuclei unit activity, responding to a velocity step in darkness, has a time course similar to that time course of the induced slow phase eye velocity, not to that of activity in the eighth nerve (Fig. 4A)(17). This indicates that the velocity storage integrator had altered the character of activity coming in over the eighth nerve at the very first processing station in the central nervous system. The difference between the time constant of neurons in the vestibular nerve and nuclei disappears under anesthesia (18). This indicates that it is important to study information processing in alert individuals if a true picture of vestibular function and of visual-vestibular interaction is to be obtained.

The velocity storage mechanism not only contributes to vestibular nuclei neuronal activity during vestibular stimulation, it also makes a striking contribution during optokinetic stimulation (17). There is a strong effect of vision on the activity of many vestibular nuclei neurons that are only one synapse away from the peripheral vestibular receptor. An example is shown in Figure 4B. Not only is there an increase in activity of this neuron during full field rotation, but also persistence of activity during OKAN (Fig. 4B). Activation during rotation in light (Fig. 4C), corresponds closely to eye velocities generated in response to the same stimulus (Fig. 2C).

Since frequencies of neurons in the vestibular nuclei are related to eye velocity during OKN and OKAN (Fig. 4)(17, 19, 20), it would seem reasonable that velocity storage is realized in the vestibular

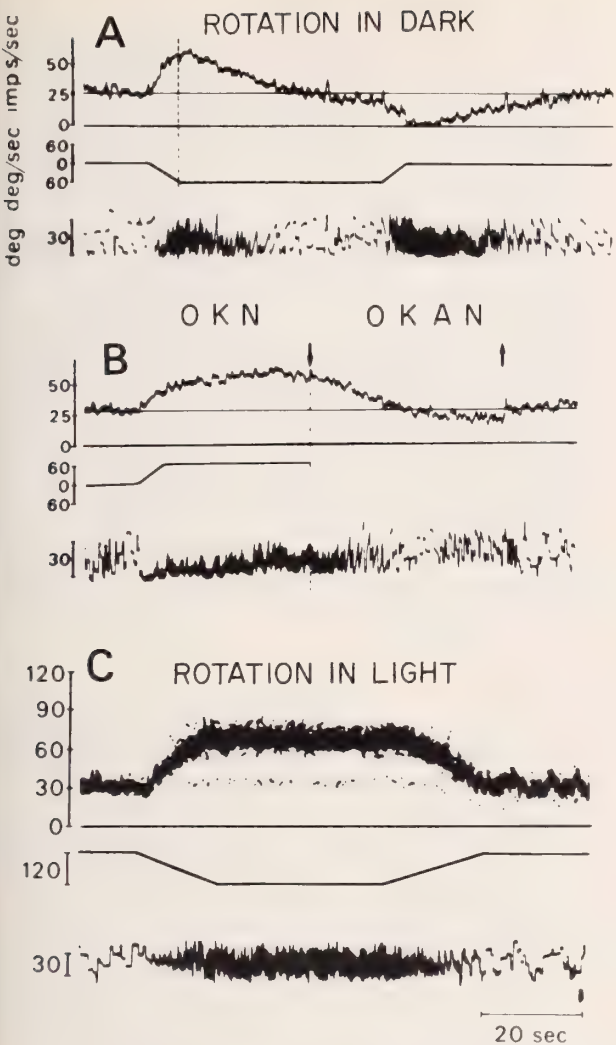


FIG. 4. Extracellular activity of a neuron in the vestibular nucleus in response to a step in angular velocity of the animal in darkness (A), of the visual surround (B), and of the animal in a stationary lighted surround (C). Note the similarity of these curves to the slow phase velocity envelopes shown in Fig. 2 and to the model stimulations shown in Fig. 3.

nuclei. However, this is probably not the case. During OKN, firing rates in the vestibular nuclei approximate the state of the integrator up to saturation levels of only about 60 deg/sec (17). The integrator can be charged by OKN to 90–120 deg/sec (12). Moreover, vestibular nuclei unit activity during rotation in dark agrees more closely with slow phase eye velocity than with activity predicted for the integrator (Compare Fig. 3A, 3B and Fig. 4A with the integrator trace shown in Fig. 3F). This suggests that the signal in the vestibular nuclei represents a summation of activity from the direct vestibular pathway and the integrator. Both the state of the integrator and the direct visual pathway are probably located elsewhere.

Other data support this view. Waespe and Henn

(19) compared firing rates during rotation in dark (Fig. 5A, upper trace) to firing rates during rotation in light in which there was no relative movement between the monkey and the visual surround (Fig. 5A–5C, lower trace). In the latter condition, the relative stationary surround was achieved by rotating the animal together with its visual environment, the OKN drum, at the same velocity. This is a “conflict” situation. That is, the vestibular system signals to the animal that it is moving, but the visual system indicates that it is stationary. At low accelerations (Fig. 5A), unit activity in dark (upper trace) achieved higher frequencies of firing than during rotation in a relative stationary surround (lower trace). As the acceleration increased, the unit activity in a relative stationary surround achieved peak frequencies that were closer to those found during rotation in dark (Fig. 5B, 5C). In all cases the eye velocity was approximately zero.

UNIT ACTIVITY MODEL PREDICTIONS

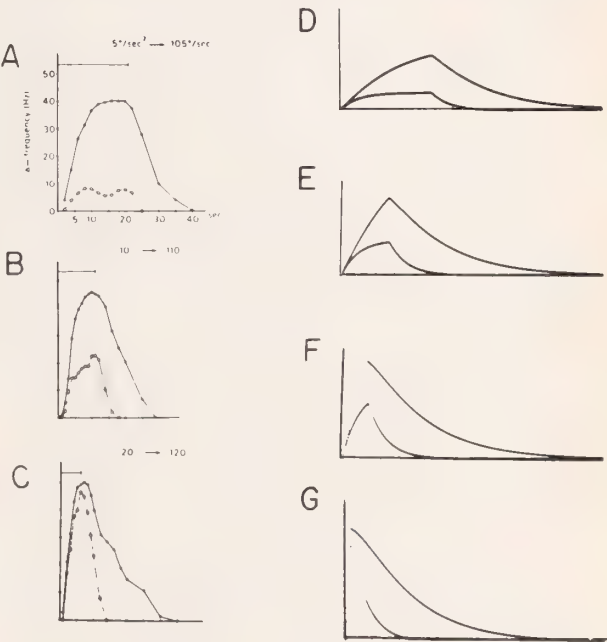


FIG. 5. Comparison of unit activity recorded in medial vestibular nuclei with activity predicted by the model. The upper trace shows the neural response to rotation in dark, the lower trace the response to rotation in light in a head-fixed surround. A, At low accelerations (5 deg/sec/sec), unit activity in the dark (upper trace) reached higher frequencies than unit activity during rotation in a stationary surround relative to the monkey (lower trace). B–C, Increasing rates of acceleration of rotation (10 deg/sec/sec and 20 deg/sec/sec) induced frequencies of firing in the stationary surround that were closer to frequencies of firing in the darkness. D, E, In all cases, eye velocity was approximately zero. F, Model predictions of vestibular nuclei unit activity for increasing accelerations of rotation in dark (upper trace) and in a lighted stationary surround relative to the subject (bottom trace).

If one considers the summation of activity from the direct vestibular pathway and the integrator as the variable related to vestibular nuclei unit activity, the model predicts the vestibular nuclei unit activity of Waespe and Henn. The experiment shown in Figs. 5A-5C was simulated in Figs. 5D-5G. In each case the lower trace represents unit activity that would be expected for rotation in light with a head fixed stationary environment and the upper trace the response during rotation in dark. The similarity of the experimental and model data indicates that the organization of the VOR may be close to the scheme that has been proposed.

Conclusions

Evidence is presented to show that integration is a basic process that is used in the realization of the vestibulo-ocular reflex. There is a mechanical integration in the peripheral labyrinth and several neural integrations in central structures. The mechanical integration acts to convert head acceleration to a signal related to head velocity. The neural integrator that has been described in the VOR responds to the incoming information and outputs to the vestibular nuclei. The consequence is that unit activity has a longer time constant than the activity coming in over the vestibular nerve. A third neural integrator converts eye velocity to eye position (11). These integrators are of fundamental importance in producing slow phases of vestibular nystagmus, OKN and OKAN. The neural integrator that converts activity with a short time constant in the vestibular nerve to a longer time constant in the vestibular nuclei has the time constant of both the behavioral response to movement, nystagmus, and under certain circumstances to perceptual responses (21). Using these integrators, it has been possible to model the vestibulo-ocular reflex and visual-vestibular interactions. The validity of this approach for studying the organization of the vestibular system is shown by the close correspondence of variables in the model to unit activity.

One of the major functions of the VOR is to compensate for head movement. The VOR acts to hold the angle of gaze stable in space. The stored activity that has been demonstrated in the VOR helps produce better ocular compensation in several ways. First, it lengthens the time constant of the vestibular response so that the VOR has a better low frequency response in the absence of vision. The gain of the VOR is improved for slow head movements, and the response is closer to being compensatory. Second, the stored activity smooths and maximizes the velocity of the eyes during the

ocular response to optokinetic stimulation. Third, the stored activity provides a mechanism whereby after-responses to prolonged rotation are abolished or are markedly attenuated. Finally, the storage mechanism serves as a focus for establishing visual-vestibular interactions, and plays an important role in helping the vestibular system generate a signal in the vestibular nuclei that more closely represents movement of the organism relative to its external surround. This is the signal that the organism requires to respond to motion. To generate appropriate eye and postural responses, it is important to know not only when one starts and stops moving, but whether one is continuing to move. Such information could come from the mechanism that is described. How it might be deranged after lesions of the peripheral or central vestibular system and how this derangement might contribute to vertigo and abnormal sensation of movements is a subject for future research.

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Student's Corner

Paul Celano, B.S., Editor

Grand Multiparity Associated with Unilateral Renal, Ovarian, and Müllerian Agenesis

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Abstract

A case of unilateral Müllerian, ovarian and renal agenesis, which was discovered accidentally at the time of surgery in a fifty-year-old grand multiparous female, is presented. Grand multiparity in patients with congenital Müllerian anomalies is infrequent. A review of the literature is presented.

Introduction

Congenital Müllerian anomalies have been diagnosed very infrequently in grand multiparous women. Unilateral Müllerian agenesis has been reported to be associated with the highest pregnancy wastage rate among all forms of uterine anomalies. The case presented here indicates that fetal wastage in cases of unilateral Mullerian agenesis may not be as common as has been suggested.

Case Report

M.T., a 50-year-old Hispanic female, G11 P11-0-0-9, was admitted to the Department of Obstetrics and Gynecology of the Mount Sinai Medical Services at Elmhurst General Hospital, Queens, N.Y.,

with a six-month history of perimenopausal bleeding. Her 11 pregnancies were followed by normal, spontaneous vaginal deliveries, with the last occurring at age 34. Additional complaints at the time of admission included pelvic pressure and urinary stress incontinence. A Pap smear was negative for malignant cells and a dilation and curettage revealed severe adenomatous hyperplasia. The patient was advised to undergo hysterectomy.

Review of systems, family and social histories were noncontributory. The physical examination was within normal limits. The pelvic exam revealed a large cysto- and rectocele and a second degree uterine descensus. The uterine cavity was approximately 8 cm in depth and the adnexa were normal. On admission, hemogram, white cell count with differential, SMA 6 and 12 were all within normal limits. Electrocardiogram and chest x-ray were also normal.

At the time of vaginal hysterectomy, absence of the left ovary and fallopian tube were noted. A right salpingo-oophorectomy and anterior-posterior colporrhaphy with Kelly application were performed. The patient's postoperative course was unremarkable.

* This work was completed during the medical student's fourth-year.

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An intravenous pyelogram obtained postoperatively revealed an absent left kidney and ureter.

Embryology

In human and mammalian embryos, the primordial germ cells migrate to the region of the genital ridges in the fourth week of development. One week later, the indifferent gonad begins to form and incorporates the primordial germ cells into its structure (1).

Three different but slightly overlapping kidney systems are formed during intrauterine life. The pronephros, which is the first to form, begins to regress before its entire structure is formed. Only the pronephric duct remains to be incorporated into the next kidney, the mesonephros, which appears at four weeks gestational age. From then on, it is referred to as the mesonephric or Wolffian duct and, in the male, will represent the conduit for germ cells from the testes to the exterior (1).

The gonad, with its primordial germ cells and the mesonephros, are suspended from the dorsal body wall by a common mesentery and are jointly referred to as the urogenital ridge (1). Normally the growth of the gonad continues but the mesonephros degenerates and is replaced by the metanephros which will form the permanent kidney.

The Müllerian, paramesonephric, or female reproductive duct arises de novo lateral to the mesonephric duct in the sixth week. About the tenth week, either the Müllerian or Wolffian duct will degenerate, depending upon the absence or presence of H-Y antigen and a fetal testis. The H-Y antigen, a Y-linked histocompatibility antigen, is a cell surface protein through which the Y-chromosome directs the indifferent embryonic gonad to organize a testis instead of an ovary (2). Although

H-Y antigen is not of itself sufficient to guarantee masculinization of the indifferent gonad, its presence seems necessary for normal testicular differentiation to occur. However, the absence of H-Y antigen does not always signal normal ovarian differentiation (3). The differentiation of the Müllerian duct into female structures, is not dependent upon the presence of an ovary. The distal Müllerian ducts on either side fuse in the midline in the ninth week with subsequent disappearance of the central wall between them. The unfused, cephalic part of the ducts become the fallopian tubes. The caudal, fused part attaches to the urogenital sinus which, beginning at twenty weeks, canalizes longitudinally to form part of the vagina (1). Cessation at any point during Müllerian differentiation and subsequent canalization can produce a wide variety of anomalies (Table I).

Renal Agenesis

Congenital absence or dysgenesis of the kidney, occurs, according to a variety of authors, in 1/500 to 1/1000 individuals (4, 5). Several theories have been proposed to explain the lack of kidney maturation in the human fetus. Allen (4) hypothesized that a deficiency of the metanephros might result in an inability of the ureteric bud (precursor of the ureters) to induce differentiation of the metanephros into the definitive kidney. Radasch (6) suggested that renal agenesis might be due to regression of the ureteric bud or to failure of the pronephros and mesonephros to form, with the consequent failure of the ureteric bud to develop. Ashley and Mostofi (7) classified renal agenesis and dysgenesis into six groups (Table II) based upon their hypothesized morphogenetic mechanism for the specific defect. For the purposes of this discussion,

TABLE I
Combinations of Uterus, Cervix and Vagina Triad*

Uterus	Cervix	Vagina
1. Single uterus	Single	Single
2. Single uterus	Septate	Septate or double
	Double	Single, septate or double
3. Septate uterus (uterus septus)	Single or septate	Single, septate or double
	Double	Single, septate or double
4. Bicornuate uterus	Single or septate (uterus bicornis unicolis)	Single, septate or double
	Double (uterus bicornis, bicollis)	Single, septate or double
5. Double uterus (uterus didelphys)	Double	Single, septate or double
6. Unilateral maturation of uterus (uterus unicornis) with or without a rudimentary horn.	Unilateral maturation of cervix	Single

*Adapted from Obstetric Significance of Genital Anomalies, Jones W, Obstet Gynec 1957; 10:113-127.

TABLE II
Proposed Classification of Renal Defects*

Type	Mechanism	Result (summarized)		Wolffian Duct
		Kidney	Ureter	
1	Failure of nephrogenic ridge to show any potentiality for differentiation as renal tissue.	Absent	Absent	Absent
2a	Metanephric blastema fails to develop and induction of the formation of a ureteric bud is absent.	Absent	Absent	Present
2b-i	Stimulus for formation of ureteric bud is present but ability of lower end of urogenital ridge to form renal tissue is completely deficient.	Absent	Present	Present
2b-ii	Same as in 2b-i, but ability of lower end of urogenital ridge to form renal tissue is partly deficient.	Dysgenetic	Present	Present
2c	Nephrogenic potentiality of lower most part of the urogenital ridge is retained, but ureteric bud fails to develop for various reasons.	Dysgenetic	Absent	Present
2d	Stimulus for formation of ureteric bud is received from opposite side of body, the ureter is attracted to that side, resulting in a double renal pelvis.	Absent	Present on opposite side.	Present

*Adapted from Renal Agenesis and Dysgenesis, Ashley D, Mostofi F: J Urol 1960, 83:211-230.

only Type 1 will be considered. In Type 1 defects, the potential for organogenesis by the nephrogenic ridge fails to appear and results in the absence of all structures derived from this area. Thus the pronephros, mesonephros and Wolffian duct will not form and the metanephros will not form the definitive kidney. Their study, as well as others conducted by Daro (8) and Collins (9) revealed a more common involvement of the left side in the cases of unilateral renal agenesis. Of 232 cases with unilateral agenesis reported by Ashley and Mostofi (22 of whom were female), the left side was involved in 132 (7). Daro (8) reported two cases of unilateral agenesis both occurring on the left side. Collins (9) reported 581 cases of renal agenesis 231 of whom were female and 69 in which the sex was not stated. Among the 581 patients, the left kidney was absent in 318 cases and in 25 the side was not reported. The ureter was absent on the side on which the kidney was absent in 297 cases and, of these, the left kidney was absent in 182 cases.

In a few cases of Type 1 defect, the area of embryonic deficiency may extend to the urogenital ridge resulting in deficient development of the gonad as well as the kidney. This was the case in the patient presented in this paper. Only three among the 22 females with unilateral renal agenesis in Ashley's series had, in addition, complete absence of one ovary, absence of the fallopian tube on the same side as the ovary, and a hemiuterus.

Müllerian Agenesis

Of the three principal groups of genital anomalies in the female, the most common are those resulting

from incomplete fusion of the Müllerian ducts (10). Unilateral maturation of the Müllerian system, with absent or incomplete development on the opposite side, results when the Müllerian duct on one side fails to develop or to reach the urogenital sinus. This type of genital anomaly appears to be most frequently associated with absent or abnormal renal structures (10).

Cytogenetic studies of individuals with Müllerian agenesis usually show a normal 46XX karyotype (16, 17). However, chromosome abnormalities of still unknown significance have been detected. A series by Sarto (18) reported two patients with uterine and vaginal aplasia. Both had chromosome abnormalities, one a 46XXt (10q⁺, 12q⁻) and the other a 45X/46XX/47XXX chromosomal constitution.

Combined Renal and Müllerian Agenesis

The association of renal and female genital anomalies has been recorded before by Radasch (6) and Collins (9). Of the 231 females in Collins' series with unilateral renal anomaly, there was an associated genital anomaly in 89%. Woolf and Allen (14) reported four cases of uterus unicornis in which three lacked one kidney on the side of the Müllerian agenesis and the remaining case had a pelvic kidney. Renal anomalies are reported to be present in about 40% of cases of Müllerian aplasia and vertebral anomalies occur in about 10% of cases (19, 20). The most common renal anomalies are unilateral agenesis and ectopic kidney (21).

It has been suggested that the defects of the Müllerian system, ovaries and kidney have the same

embryologic cause (7). It was noted earlier that the Wolffian and Müllerian ducts develop in close anatomical relationship. It has been suggested that a defect in the entire region of the urogenital ridge could account for failure of multiple organ development. Gruenwald (22) demonstrated in 1941 that section of the Wolffian duct resulted in an absent kidney and fallopian tube and an unicornuate uterus. This was lack of development of the metanephros and the ureteric bud. Such a condition was present in this case and thus reinforces the importance of an urologic work-up in the presence of Müllerian abnormality to detect renal agenesis, dysgenesis or ectopy or vice versa.

Discussion

Although congenital anomalies of the female reproductive tract are encountered infrequently in obstetric practice, they are of concern because of their association with increased frequencies of abortions, premature labor and malpresentations (11-13). Woolf and Allen (14) reported the incidence of these anomalies to be 1/500 to 1/600 women or 1/2000 gynecologic cases. About 20 years ago, efforts were made to correlate the obstetric performance with the type of anomaly. The significance of these anomalies, according to Jones, is directly related to the volume and contractility of the uterine muscle mass (10). A hemiuterus, the result of unilateral Müllerian maturation or complete lack of Müllerian fusion, should give the worst obstetric performance of all uterine anomalies (10). Because of its reduced muscle volume its ability to expand and hypertrophy is diminished compared to that of the normal uterus. Furthermore, the hemicervix accompanying the hemiuterus should be limited in its ability to dilate because of its inadequate structure (10). This theoretical concept was reinforced by subsequent similar studies (11, 12, 23).

Of the 123 women with genital tract anomalies studied by Jones (10), sixteen were reported to have a hemiuterus. Four women had unilateral Müllerian agenesis (uterus unicornis) while 12 had complete lack of fusion of the Müllerian ducts (uterus didelphys) which is the equivalent of two hemiuteri. Thirty-seven pregnancies were reported among these 16 women, of which 32.4% resulted in spontaneous abortion and 60% in fetuses carried to 38 weeks or more (10). Breech presentations occurred in 40%, and 72% of the viable pregnancies required primary cesarean sections, reflecting the inefficiency of the hemicervix (10). Including abortions, the gross fetal wastage was 46% (10).

The largest series reported specifically correlating uterus unicornis with obstetric performance is the one by Hay (15). He reported eight cases of uterus unicornis, six of which had an absent left horn. The eight cases resulted in 14 pregnancies, six of which aborted spontaneously. However, none of the remaining eight went into premature labor, the reason for which was unexplained by Hay. The total fetal wastage was nine out of 14 pregnancies. Six of the eight term pregnancies were breech presentations. Three of these eight patients had an infertility problem.

The patient presented here is most unusual because of the simultaneous occurrence of renal, Müllerian and ovarian agenesis, in association with grand multiparity. In spite of her Müllerian abnormality, which has been associated with the worst possible obstetrical performance (10), the patient carried 11 pregnancies to term without complications.

Conclusion

A grand multipara with unilateral ovarian, Müllerian and renal agenesis has been presented followed by a review of the literature. She has demonstrated that fertility may not be impaired in the presence of Müllerian agenesis despite literature which supports the belief that its obstetric performance is the worst of all the genital tract anomalies. Premature labor, spontaneous abortion and breech presentations apparently occur more frequently with these anomalies but because of this patient's uneventful obstetric history, no uterine anomaly had been suspected. The actual incidence of congenital Müllerian abnormalities may therefore be higher than reported. The reported high incidence of pregnancy loss in association with a hemiuterus may be exaggerated due to a bias in patient selection in published series. Most uterine anomalies are unsuspected until obstetrical complications arise. Patients without complicated pregnancies and deliveries may go undiagnosed unless some other problem warrants evaluation of the genital tract. Complications may arouse one's suspicion and subsequently lead the obstetrician, via hysterosalpingogram, to make a diagnosis of unilateral Müllerian agenesis. One cannot conclude with certainty that any or all complications arose as a consequence of the agenesis. However once the diagnosis is made the obstetrician is influenced to ascribe any spontaneous abortion, premature labor or malpresentation to the patient's anomaly. Thus repeated pregnancy wastage may have resulted in an inappro-

priate representation of "high risk" cases in published series.

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Patterns of Injuries in College Athletes: A Retrospective Study of Injuries Sustained in Intercollegiate Athletics in Two Colleges over a Two-year Period

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Abstract

A retrospective study of sports related injuries incurred at two New York City Colleges and referred to The Mount Sinai Hospital Sports Medicine Program (MSH-SMP) over the past two years was undertaken. During the study, 1,142 students participated in twenty-two sports and sustained one-hundred and seven injuries, an injury rate of 9% per year. Contact sports, notably wrestling, male basketball and soccer, had an injury rate twice that of noncontact sports, notably golf, softball, male swimming, tennis and volleyball. An exception was female gymnastics, which presented with the highest injury rate of all teams. Sprains (28%) and strains (19%) were the most common injuries. Nearly one half the injuries involved the lower extremity, with the knee (24%) and ankle (15%), the anatomic sites most often involved. Of the one-hundred and seven injuries, 48% were seen at The Mount Sinai Hospital Sports Medicine Program for evaluation and treatment. Injury data for the referred population closely approximated that found for the total injured population as a whole.

Introduction

During the past twenty-five years, America has witnessed a virtual explosion in sports participation. From the Little League baseball player to the elderly jogger, sports have become an integral part of the daily lives of millions of Americans. However, paralleling this popularity has been a steady increase in athletic injuries. A recent government survey estimated that more than one million injuries occurred in high school and college athletic programs during the 1975-76 school year. The report revealed that football is by far the most hazardous major sport, with an injury rate four times that of noncontact sports. The injury rates in other contact sports (for example baseball, basketball, football, hockey, lacrosse, soccer and wrestling) were 74 per 1,000 for men and 54 per 1,000 for women. In noncontact sports (bowling, crew, cross country, golf, swimming, tennis and track) the rates

were 38 per 1,000 for men and 32 per 1,000 for women. In response to these nationwide statistics, former HEW Secretary, Joseph Califano, commented that, "the casualties may have reached unacceptably high levels" (1).

At present, a large volume of literature exists on the treatment of athletic injuries. Prevention, only recently, has started to assume a comparable level of information. The general health of the athlete, his/her physical condition and motivation are all crucial in determining the risk of injury. In addition, the type of sport, its rules and regulations, the playing surface, the equipment utilized and the coaches and trainers also play an important role in the incidence and nature of injuries. Now that sports medicine has become a significant discipline in the medical field, information on prevention of athletic injuries is growing rapidly.

There is still a paucity of data on injury rates and classification of injuries for specific sports (2-11). Such data is a basic requirement for the evaluation of what changes are needed to enhance the safety of the athlete. The purpose of this paper is:

- 1) To study the incidence and nature of injuries

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in a representative population of athletes engaged in organized collegiate sports.

- 2) To compare the total injured population with those injuries referred to The Mount Sinai Hospital Sports Medicine Program (MSH-SMP).
- 3) To discuss the importance of an athletic trainer.

Materials and Methods

A retrospective study of all sports related injuries incurred at two New York City colleges and those injuries referred to the MSH-SMP during the 1977-78 and 1978-79 academic years was conducted by a review of: 1) rosters of participants in all sports played, injury reports, and injury referral lists to the MSH-SMP obtained from the athletic departments of both colleges, and 2) the MSH-SMP injury reports and outpatient charts of all injuries seen at the hospital. Reported injuries were recorded by the trainer at each college either soon after they occurred or after they were evaluated and treated at the MSH-SMP, as several injuries were sent directly to the hospital before being seen by the trainer. Injury diagnoses were made by the trainers and by the MSH-SMP physicians for those injuries seen at the hospital. Injuries were defined as any disability resulting from intercollegiate sports participation for which medical attention was sought or required, regardless of whether it occurred during practice or competition.

The colleges chosen for this study maintained an exclusive affiliation with the MSH-SMP. The MSH-SMP functions as an outpatient clinic specializing in evaluation and treatment of all sports related injuries. The study population consisted of all students from the two colleges participating in organized intercollegiate sports during the two-year study period. With the exception of football, not played at either college, most major sports were represented.

Results

Injury Data for Total Injured Population

Injury Rates (Tables I, II): During the study period, a total of 1,142 students participated in 22 sports and sustained one-hundred and seven injuries, an injury rate of 9.0%. The rate of males was 8.9%, while for females it was 9.6%. Eighty-two percent of the females participated in noncontact sports, whereas male participants were evenly represented between contact and noncontact sports,

55.0% and 45.0% respectively. In noncontact sports, women had an injury rate of 8.9% and men had a rate of 4.5%. In contact sports the female rate was 8.6%, while for males it was 13.9%.

Female gymnastics, a noncontact sport, had the highest injury rate, 30.0%, off all teams. Wrestling, male basketball and soccer, all contact sports, had the next highest injury rates, 19.1%, 18.8%, and 16.9% respectively. Hockey, although it had the highest injury rate among contact sports, was not considered representative since it was only played for one season at one school. At the other extreme, six teams had no injuries at all, those being golf, volleyball, softball, female and male tennis and male swimming.

TABLE I
Injury Rates (Total)

		No. of Participants	No. of Injuries	Injury Rates %
All	Total	1,142	107	9.4
Sports	Female	384	34	8.9
	Male	758	73	9.6
Contact Sports	Total	476	63	13.2
	Female	58	5	8.6
	Male	418	58	13.9
Noncontact Sports	Total	666	44	6.6
	Female	313	28	8.9
	Male	353	16	4.5

TABLE II
Team Injuries (Total)

Sports	No. of Participants	No. of Injuries	Injury Rates %
Baseball (M)*	72	4	5.6
Basketball (F)*	36	5	13.9
Basketball (M)	101	19	18.8
Cheerleaders (F)	35	2	5.7
Fencing (F)	55	2	3.6
Fencing (M)	32	2	6.3
Golf (M)	10	0	0.0
Gymnastics (F)	30	9	30.0
Gymnastics (M)	21	3	14.3
Hockey (Ice) (M)	13	4	30.8
Lacrosse (M)	42	3	7.1
Soccer (M)	65	11	16.9
Softball (F)	58	0	0.0
Swimming (F)	45	6	13.3
Swimming (M)	39	0	0.0
Tennis (F)	34	0	0.0
Tennis (M)	37	0	0.0
Track (F)	92	9	9.8
Track (M)	214	11	5.7
Volleyball (F)	22	0	0.0
Wrestling (M)	89	17	19.1
Totals	1142	107	9.4

M = male

F = female

Injury Type (Table III): Sprains (28%) and strains (18.7%) accounted for nearly one half the total injuries. Contusions (11.2%) were the next most common injury followed by lacerations (5.6%).

Injury Location (Table IV): The lower extremity was involved in one half of all injuries, with the knee (24.3%), ankle (15.0%) and thigh (5.6%) being the most commonly involved sites. The upper extremity was involved in 18.7% of all injuries, with the shoulder (7.5%) the most common site. Of the nonextremity locations, the head (6.5%) was most frequently involved.

TABLE III
Injury Type

Type	Total Population		Participants Seen at MSH-SMP	
	No.	%	No.	%
Contusion	13	12.2	8	15.7
Dislocation	4	3.7	3	5.9
Fracture	5	4.7	4	7.8
Laceration	6	5.6	5	9.8
Sprain	30	28.0	9	17.6
Strain	20	18.7	7	13.7
Inflammation	4 ^a	3.7	4 ^c	7.8
Other	25 ^b	23.4	11 ^d	21.6
TOTALS	107	100.0	51	99.9

a & c - arm infection (1), bursitis (1), tendinitis (2).
b - dental (2), hernia (1), hip pain (2), meniscal tear (2), ulnar nerve compression (1), patellar tendon calcific mass (1), type unspecified (16)
d - chest pain (1), hernia (1), hip pain (2), meniscal tear (2), patellar tendon calcific mass (1), ulnar nerve compression (1), charts unavailable (3).

TABLE IV
Injury Location

Location	Total Population		Participants seen at MSH-SMP	
	No.	%	No.	%
Head	7	6.5	6	11.8
Neck	2	1.9	1	2.0
Chest	3	2.8	3	5.9
Back	3	2.8	2	3.9
Shoulder	8	7.5	2	3.9
Arm	2	1.9	1	2.0
Elbow	4	3.7	4	7.8
Wrist	2	1.9	2	3.9
Hand	3	2.8	3	5.9
Finger	1	0.9	0	0.0
Hip	3	2.8	3	5.9
Thigh	6	5.6	1	2.0
Knee	26	24.3	10	19.6
Ankle	16	15.0	6	11.8
Foot	4	3.7	1	2.0
Others ^a	17 ^a	15.9	6 ^b	11.7
Totals	107	100.0	51	100.1

a - eye (2), groin (2), mouth (2), location unspecified (1).
b - eye (1), groin (2), charts unavailable (3).

TABLE V
Referrals to MSH-SMP

	College A	College B	Totals
No. Participants	526	616	1142
No. Injuries	65	42	107
No. Referred to MSH-SMP	21	37	58
No. Seen at MSH-SMP	21	30	51

Sprains accounted for more than one half of both knee and ankle injuries. Strains were responsible for three fourths of the shoulder injuries and all of the thigh injuries.

Sport Related Injuries: Among the sports with the highest injury rates, the knee and ankle were the most frequent sites affected. Wrestling injuries (17) comprised 23.5% knees and 17.6% ankles. Male basketball injuries (19) involved the knee in 31.6% followed by the ankle with 21.1%. Soccer, with 11 total injuries, was represented by 27.3% knee and 18.2% ankle.

Injury Data of Referrals to MSH-SMP

Referrals to MSH-SMP (Table V): A total of one-hundred and seven injuries occurred during the study period, of which 58 were referred to the MSH-SMP. Of those referrals, only 51 were actually seen since seven failed to come to the hospital.

College A referred only one third its total (65) injuries, whereas college B referred greater than three fourths its total (42) injuries. All of college A's referrals were seen at the MSH-SMP, whereas only 30 of college B's 37 total referrals were seen, explaining the seven referrals not seen at the hospital.

Injury Type (Table III): A trend similar to that for the total injured population is seen. Sprains (17.6%), contusions (15.7%) and strains (13.7%) were the most common injury types seen at the MSH-SMP.

Injury Location (Table IV): Again, a similar pattern emerges, with the knee (19.6%), ankle (11.8%) and head (11.8%) the most frequent sites involved. The upper extremity represented 23.5% of the injuries and the lower extremity, 39.3%.

Sprains accounted for approximately one third of both knee and ankle injuries. One half the ankle injuries were fractures.

Discussion

Injury Rates: It is difficult to compare the injury rates found in this study with those of others due to differences in athletic population, definitions of what classifies an injury and injury reporting methods.

Contact sports had an injury rate twice that of noncontact sports and this was statistically significant ($p < .01$). Such a finding is not uncommon, as other investigators have obtained similar results (2, 4, 6-8, 11). The one exception was female gymnastics, which presented with the highest injury rate of all the teams. However, Snook in 1979, also found women's gymnastics to be a very hazardous sport (5).

To compare male versus female injury rates in contact sports would be meaningless, since sports offered in the male program (soccer, lacrosse, wrestling) were not offered in the female program. In noncontact sports, however, there was a comparable distribution—females had an injury rate (8.9%), twice that of males, (4.5%) and this was statistically significant ($p < .03$). This difference can be attributed to a lack of proper skills, conditioning and coaching for females, and increased reporting of injuries for females (12).

Injury Type and Location: Strains, sprains and contusions accounted for the greatest percentage of injuries (Table III), while the knee and ankle were found to be the most frequently injured sites (Table IV). The large number of lower extremity injuries involved sports like basketball, soccer, track and wrestling in which twisting, acceleration and deceleration motions are prone to cause injuries to that area.

Only eight of the one-hundred and seven total injuries required surgery. There were six knee and two shoulder operations. Such a low number of operations is, in part, because football is not played by either college. Football, because of the necessary contact and torsional injuries to the weight-bearing joints of the lower extremity, especially the knee, is responsible for many injuries requiring surgery.

Referrals to MSH-SMP: One of the main reasons for the difference in referrals between the two colleges is college A's policy of maintaining a full-time trainer who practices triage—a system whereby all injuries are evaluated, the minor ones are treated and the more serious ones are sent to the MSH-SMP. Such a system is extremely valuable since the "major factor in the management of athletic injuries is the earliest possible determination of the type and extent of injury" (13). No injury from college A was sent to the MSH-SMP without first being seen by the trainer. At college B there was a part-time trainer who did not screen injuries because of his unavailability at the time many injuries occurred. He, therefore, referred all injuries to the MSH-SMP, regardless of severity, unless the injured athlete decided to go to a private physician or not be treated at all. Although several injuries at

college B were sent to the MSH-SMP before being seen by the trainer, he eventually saw them for follow-up rehabilitation as prescribed by the physicians at the MSH-SMP.

It can be appreciated that the trainer can play a crucial role in limiting the number of injuries sent to the MSH-SMP, injuries which could easily be treated at the college. The trainer, in addition to practicing triage, is familiar with each individual athlete and the aspects of a sport that lead to injury. He can supply information concerning conditioning and safety, helping to prevent many new injuries (14). It has been shown that those schools with trainers have appreciably lower injury rates (15).

In summary, the data presented in this paper give the reader some idea about which intercollegiate sports (football excepted) are at high risk for injury, the location and type of the most common injuries seen in the total population studied, and at The Mount Sinai Hospital Sports Medicine Program, and the value of ancillary personnel, most notably the trainer.

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Introduction to Immunology

HUGH R. K. BARBER, M.D.

Introduction

The medical world has turned over many times since I was a medical student. Some specialties have come into their own while others have faded in importance. Today, immunology is the glamor specialty of medicine cutting across all specialties.

Modern immunology started when Jenner discovered that inoculation with cowpox protected man against smallpox. The next great advancement in immunology was made by Louis Pasteur. Many contributions followed in the field of infectious diseases. Attempts to convert these contributions towards earlier diagnoses and therapy in oncology met with little success (3, 4).

The medical scientists of that time concluded that tumor specific antigens logically should be present. Proof of this concept was not forthcoming for another half a century. Numerous attempts at cancer immunotherapy have been undertaken since the turn of the century when studies with out-bred laboratory animals demonstrated that strong immunity could be induced against transplantable rodent neoplasms. There followed a period of intense laboratory and clinical investigation of tumor immunotherapy with anticipation for subsequent control of malignant disease. However, it soon became evident that the immunity demonstrable against normal alloantigens (histocompatible antigens, HL-A) was carried in tumor cells rather than against tumor specific antigens. Although an occasional striking regression occurred, the clinical use of autologous vaccines as immunoadjuvants was equally disappointing. Thus, clinical interest in immunotherapies declined as rapid advances in the fields of surgery, radiation therapy, and chemotherapy provided more promising modalities for cancer therapy.

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It is difficult to realize that during my student days at Columbia College of Physicians and Surgeons, I had the opportunity to be taught by Michael Heidelberger, one of the modern day pioneers of immunology. Alphonse Raymond Dochez was phasing out as the Chairman of the Department of Bacteriology at that time. They had been preceded by Philip H. Hiss, Frederick Parker Gay and Hans Zinsser. These scientists represented notable figures in Columbia College of Physician and Surgeons' great tradition of immunology. Although it all seemed so esoteric, impractical and far away, it obviously stimulated and challenged the minds of students who studied during that era. The school has continued its leadership in the science of immunology.

Three difficulties face the student of any rapidly developing and expanding subject. It has been difficult to establish a common vocabulary. However, this has now been fairly well achieved. The advancing frontiers always seem very complicated and usually are until they are understood and placed in their right perspective. Last in this list is the task of incorporating these advances into the clinical care of the patient. It gives relevance to the subject and permits the physician to accumulate his own experience. Immunology is the subject of the future and although it is changing almost daily, a sound background in the basic principles of the subject will allow the practitioner to grow with the subject (10, 11, 27).

Immunology is a relatively new medical discipline, the basic concepts of which are fundamental for an understanding of the principles and practice of modern medicine. Although immunology is relatively new, the concept of immunity has been known from ancient times; resistance to disease was recognized by the ancient Chinese. Not only did the Chinese recognize the concept of immunity, they were even able to identify that it had specificity and that immunity to smallpox did not protect them against other diseases. The term immunity has now

taken on an expanded role. It is defined as the property whereby the lymphoreticular system makes a memorized response to an antigenic stimulation. This may result in a state of positive reaction known as sensitization in which specific antibodies are developed to control invading and foreign antigens, or in one of the negative reactions, known variously as immunologic tolerance, or immunosuppression. The former is protective to the host and offers resistance to tumor development, viruses, intracellular bacteria, fungus and parasites. The latter plays a role in women in the protection of a pregnancy, keeping the fetus from being rejected by its host and mother. There is a continuum of immune responses from sensitization to tolerance.

The future of modern immunobiology was assured when Edward Jenner, as a medical student, made the surprising and sophisticated discovery that inoculation with cowpox cross protected man from smallpox. This important discovery resulted from Jenner's observation that milkmaids who had contacted cowpox were resistant to infection with smallpox (6).

The enhancement and further development of preventive immunization was made possible by Louis Pasteur's vaccine (vaca: cow, L. in honor of Jenner's contribution). Pasteur's researches led to the development of the germ theory of disease from which he developed techniques for the *in vitro* culture of micro organisms. This work produced material which could be used for vaccines: living, heat-killed and attenuated (living but with reduced virulence). During these investigations, Pasteur observed that all cultures (attenuated) of fowl cholera organisms inoculated into fowl produced no disease. Surprisingly, these fowl were resistant to subsequent infection with the organism and were solidly immunized. The early use of living attenuated cultures for active immunization is still the therapy of choice in the prophylaxis of many infectious diseases (28).

Héricouk and Richet felt that if this could be achieved with bacteria, the same principle should apply to the control of cancer. They injected human cancer into animals and developed an antiserum. When they later used this antiserum, the results were occasionally spectacular, but more often, the results were very poor. Some subjects developed an anaphylactic reaction, others, serum sickness. It is obvious to us now that their serum included cytolytic antibodies that could kill tumor cells but might also have enhancing antibodies that could cause the growth of tumor cells. Nor, did they realize that the

humoral mechanism requires antigen, antibody, and complement to carry through lysis.

Robert Koch, during the studies of bacterial etiology of infectious diseases, discovered the tubercle bacillus. During attempts at developing a vaccine for tuberculosis, he observed the phenomena today known as delayed hypersensitivity or cell mediated immunity. It established an additional landmark in the field of immunology.

Roux and Yersin then demonstrated the existence of an exotoxin elaborated by the diphtheria bacillus. This toxin was used by vonBering and Kitasato to inoculate animals who produced in their serum a toxin neutralizing substance called antitoxin. This neutralizing capability could be transferred by the serum to an uninoculated animal, a process called passive immunization. This work forms a model for modern techniques of prevention of disease through passive immunization (immunotherapy) (25).

Ehrlich proposed the humoral theory of antibody formation. He also hypothesized that malignant neoplasms possess antigens and, as such, are recognized as being foreign to the host. In addition, in 1908 Ehrlich indirectly suggested the theory of immunologic surveillance. He stated, "that in the enormously complicated course of fetal and post fetal development, aberrant cells became unusually latent because of the organism's positive mechanism which," he implied "was immunity." Ehrlich also theorized that a depressed immune response allows the rapid, parasitic growth of neoplastic cells. Little more was done in the pursuit of the tumor antigen from the turn of the century until 1943 when Gross reported his work using pedigreed mice.

At the beginning of this century, the hope was held that dissimilarities between normal and neoplastic cells could be demonstrated by immunologic methods, making vaccination against cancer a possibility. This concept was based on experiments in which the growth of transplanted animal tumors could be prevented by immunization of recipients of the transplants in various ways. These experiments, however, were performed with tumors originating and tested in non-inbred (out-bred) animals. The graft recipients therefore rejected the neoplastic cells not only because of an immunity to (hypothetical) tumor specific antigens, but also—and more importantly—because of sensitization to those normal alloantigens that were present in the animal in which the transplanted tumor originated and, therefore, in the tumor (but not in the recipient).

When it was recognized that the many claims of

tumor specific antigenicity were based on experiments in which an immunity to normal alloantigens rather than tumor specific antigens had been demonstrated, the field of tumor immunology came into disrepute. A common view was that the tumor specific antigens could not possibly exist, since they would confer a selected disadvantage on the cells that carry them and lead to their immediate elimination by immunologic mechanisms. That neoplasms often metastasize to lymph nodes was viewed as additional indication that an immunologic defense against cancer could hardly play a role.

In 1943, working with so called pedigreed mice which were truly inbred strains of mice, Gross was able to show that mice immunized by intradermal inoculations of tumor cells rejected a subcutaneous transplant of the same tumor, while nonimmunized animals did not. His experiments were done on very small numbers of animals, and there were few controls so that his work was largely ignored. In 1957, Prehn and Main showed that mice immunized against syngeneic methylcholanthrene-induced fibrosarcomas by inoculation of living sarcoma tissue, followed by surgical removal of the growing tumors, were resistant to subsequent grafts of the same tumor. They also showed that immunization with normal tissue did not confer resistance to the tumor graft and that mice that had become resistant to the tumor still accepted skin grafts from syngeneic mates. This work established that there was a tumor antigen present on the surface of the tumor cell.

Scientific Discipline

As a scientific discipline, immunology encompasses *immunity*, dealing with the adaptive response to infective agents; *immunochemistry*, concerned with the chemical nature of antigens and antibodies; and *immunobiology* which deals with the activity of the cells of the immune system and their relationship to each other and their environment.

Transplantation of Tissue

The terms autograft, syngeneic graft, allogeneic graft, and xenogeneic graft are defined as follows: *autograft* is a transplant in the same individual; a *syngeneic* graft (isogeneic) relates to the same inbred strain or identical twins; and *allogeneic* graft (homograft) means that the graft is from the same species but from a host with a different genetic

constitution. The *xenogeneic* graft or heterogeneic graft relates to a different species.

Antigens Present on Human Tumor Cells

Each cell has a variety of antigens present on its surface. These antigens vary from time to time depending upon various conditions that are present. On all nucleated cells, there is a human leukocyte antigen (HL-A) which identifies the host as a unique individual, and is also the antigen that stimulates an antibody response resulting in rejection of organ or tissue grafts from one individual to another who are not syngeneic (genetically alike). As each individual starts from a single cell that has to have everything present that will be needed for the rest of that patient's life, it is not surprising to find that among males and females with a variety of tumors, using highly sensitive pregnancy tests, approximately 40 percent give a positive pregnancy test; and in embryonal tumors may reach nearly 100 percent. Obviously, there are genes that have been repressed from embryonic life; some stimulus causes the genes to be derepressed thereby producing the chorionic gonadotropin antigens to appear on the surface of the cell. The work of Gold has confirmed this observation; indeed, it was demonstrated that cells that contain carcinoembryonic antigen also contain the HL-A antigen of that particular host. The HL-A antigen is a much more potent antigen and is the tumor antigen, and stimulates an immune response to a greater degree than do the tumor antigens that are present on the surface of the cell.

Antigen-antibody Interrelations

Substances that can be bound by antibody are called *antigens*. *Immunogens* are defined as substances capable of eliciting an immune response. *Haptens* are defined as antigens that, by virtue of their small size, are incapable of stimulating an immune response. Like haptens, *tolerogens* contain antigenic determinants that do not stimulate the formation of antibodies or specifically sensitized cells. Unlike haptens, tolerogens render an individual specifically unresponsive to those determinants, even when later exposed to them in a highly immunogenic form.

Tolerance is defined as a state of specific immunologic unresponsiveness to antigen that would normally induce a response. Tolerance to an antigen implies unresponsiveness towards that specific antigen while overall immunologic responsiveness to

other antigens remains intact. The most important example of tolerance is an individual's specific unresponsiveness towards self antigens. If this self tolerance state lapses, an individual mounts an immune response against his or her own cells and tissues, resulting in an autoimmune disease. Normally, individuals are immunologically unresponsive or tolerant to self antigens. Therefore, tolerance is a state of specific immunologic unresponsiveness to an ordinarily immunogenic stimulus.

Cellular Events in an Immune Response

A knowledge of the cellular events in an immune response is fundamental to understanding the basic mechanisms relative to the development of both cell-mediated and humoral immunity.

Antibody is an end product in the activation of the immune response. Antibodies, like other proteins, are composed of amino acids and synthesized intracellularly on ribosomes. Enzymes bind substrates and thus catalyze a chemical reaction; antibodies bind antigen and cause them to be removed by the reticuloendothelial system. The ability of a molecule to elicit immune response is called immunogenicity. The substance that possesses the quality of immunogenicity is called an immunogen. It is possible for a substance to be antigenic but not immunogenic. Some substances are capable of binding to antibody but themselves are incapable of inducing antibody production. A substance that is immunogenic is also always antigenic. Such substances that are antigenic but not immunogenic are called haptens. Haptens are usually small molecules (molecular weight less than 1,000 daltons) that are incapable (by themselves) of activating the cellular machinery of the immune response.

Immune Response

The macrophage is the keystone in the development of an immune response. The antigen (any substance foreign to the host) is processed by the macrophage, a mononuclear phagocyte, if an immune response is to occur. The antigen or immunogen is now a processed antigen or immunogen. Following contact of the antigen or immunogen with the macrophage, the latter sends out a message in the form of a RNA or RNA-antigen or immunogen complex. Uncommitted lymphocytes are transformed into lymphoblasts; the transformed lymphoblasts give rise to sensitized lymphocytes as well as to plasma cells. The plasma cells in turn produce a variety of immunoglobulins, five of which have been

identified: IgM, IgG, IgE, IgA, and IgD. This constitutes the humoral immune mechanism. Sensitized lymphocytes proliferate into so called killer lymphocytes that have the ability to destroy cancer cells, produce lymphokines, and produce delayed hypersensitivity. These sensitized lymphocytes constitute the cellular immune mechanism, cell-mediated immunity (CMI).

The Immune Arc

The immune system is protected by virtue of its ability to contain, inactivate, and destroy potentially harmful substances. The immune system has two means for responding to an antigen: the antibody response and the cell-mediated response. The cells that form the basis of the immune response, whether antibody mediated or cell mediated, are called lymphocytes. The cell-mediated immunity is provided by a particular subpopulation of T lymphocytes. Such cells respond to antigen by proliferating and differentiating into specifically sensitized cells (effector, or killer T cells) which, on a second encounter with the same kind of antigen, induce an inflammatory reaction in the area of confrontation. A B lymphocyte has thousands of antibodies on its surface. This cell has one immunogen specificity. Immunogen activation of a B lymphocyte induces the cell to proliferate and synthesize antibody. The specificity of the antibody that a cell secretes is the same as the antibody on its surface. The immunogenicity depends upon several factors: 1) the degree of dissimilarity between the antigen and the individual's own molecules, 2) the quantity of the immunizing dose, 3) the route of administration, and 4) the size of the antigen. Considering the inability of haptens to elicit an immune response, the best immunogens are large molecules.

The Three Phases of the Immune Arc

The immune response is divided into three phases. The first phase begins with the administration of the antigen and ends when the antigen encounters antigen reactive lymphocytes; this phase is termed the *afferent limb*. The afferent limb is the means by which the stimulus, that is the antigen or immunogen is delivered to the *central processing mechanism*, which, in turn, manufactures the effectors of the immune response, that is, antibodies and specifically sensitized cells. The dissemination of the effectors throughout the body defines the *efferent limb* of the immune reflex arc.

The Differentiation of B Cell and T Cell Immunity

Thymus dependent cells (T cells) and thymus independent cells (B cells) probably arise from a common stem cell in the bone marrow. The exact mechanism by which each cell is processed at the central level to form a T or B cell has not been completely elucidated. The differences in opinion about the origin of each cell type is probably not germane to the present discussion. However, after these cells are processed at the central level, the T cells are long lived up to years, are mobile, do not have immunoglobulin receptors, and are not acted upon by complement. The thymus derived (T cells) provide protection against viruses and intracellular bacteria, play a major role in rejection of grafts and neoplastic cells, and in the control of exogenous parasites. The T cell is directly responsible for the delayed hypersensitivity reaction and immunologic surveillance. When lymphocytes come in contact with a specific antigen to which they have been sensitized, they produce lymphokines (transfer factor, lymphocytes transforming activity, migration and inhibition factor, and lymphotoxins).

The thymus independent or bone marrow derived cells (B cells) are short lived, only a few days. They make up the humoral mechanism and have immunoglobulin receptors. The B cells require a complement for their lytic action. They are cell free antibody, and their synthesis results in the immunoglobulins (IgG, IgM, IgA, IgD and IgE). These antibodies protect the organism against bacterial infections. Bacterial vaccines inoculated into the organism result in the production of immunoglobulins.

Serum proteins can be characterized by their solubility, their mobility in an electrical field, and their size. Solubility and sodium sulphate distinguishes serum albumin and serum globulin actions. In 1937, Tiselius used electrophoresis (a process of separating proteins in an appropriate matrix by their migration in an electrical field) to delineate serum proteins further into albumin, and alpha, beta and gamma globulin fractions. Immunoglobulins are the class of serum globulins that contain antibodies. Antibodies are synthesized by an animal in response to antigenic stimulus and react rather specifically in vivo and in vitro with the inciting antigen.

The immune response takes two forms: 1) a humoral response mediated by cell free fluid containing antibodies which are circulating freely in the blood stream as well as other body fluids and 2) the

cell-mediated immunity carried out by sensitized lymphocytes (1, 2).

The humoral response (B cell) is mediated by immunoglobulins. It is cell free and made up of the immunoglobulins IgG, IgM, IgA, IgD and IgE. For the cell free antibody to have a killing or cytotoxic effect, a third substance in addition to the antigen and antibody is required. This substance, called complement, is made up of several factors present in the normal serum. Without complement, antibody can bind to the antigen on a cell surface but cannot damage or kill the cell. The nature and precise function of complement has only recently been explained. The humoral response offers a defense against most bacteria and some foreign proteins. When the host is challenged with a bacterial vaccine, the response is through the humoral mechanism with the production of immunoglobulins.

The cell-mediated response (T cell) is mediated by sensitized lymphocytes. In the cell-mediated immune (CMI) response, the antibody remains an integral part of the cells that produced it and is a cell associated with antibody. Sensitized lymphocytes carrying such antibody are carried in the lymph to tumor cells where their antibody combines with the antigen determinant on the surface of the tumor cell. The cell bound antibody then becomes capable of rupturing and killing tumor cells. The mechanism for this action has not been completely elucidated. The cell-mediated response is particularly important in graft rejection and in the elimination of viruses, intracellular bacteria, and cancer cells (18-20).

There are subpopulations of T lymphocytes, that is, *helper*, *suppressor* and *tolerant*. The helper T lymphocytes stimulate immunocompetent B cells to differentiate into plasma cells, which are the main producers of antibody (immunoglobulin). The stimulated immunocompetent B cell may produce a population of cells, possibly a variant of the B cell, that are able to attack tumor cells after the tumor cells have been exposed to specific antibody. The variant B cell does not require a complement to lyse the tumor cell and it is the only B cell capable of functioning without complement. The suppressor T lymphocytes have an inhibiting effect of the immune mechanisms. Reports indicate that autoimmunity may arise whenever there exists a state of immunologic imbalance in which B cell activity is excessive and suppressor T cell activity is diminished. A central mechanism in this concept involves a disturbance of the delicate balance between suppressor and helper activity of regulatory T cells.

Specifically sensitized T lymphocytes produce cell-mediated immune reactions by virtue of two characteristics: 1) Through their cell surface receptors, they engage antigens on the surface of foreign target cells and induce target cell destruction directly. 2) Activated T lymphocytes release into their environment pharmacologically active substances called lymphokines which promote inflammatory reactions and mediate nonspecific destruction of other cells in the immediate environment. Lymphokines are secreted by antigen activated T lymphocytes and lack specificity for antigen.

Burnet's Clonal Selection Theory

Burnet's clonal selection theory has become the central dogma of immunology. The immune system is capable of responding to a wide variety of naturally occurring and synthetic immunogens. This diversity of recognition and response is provided by the sum of different specificities of the entire lymphocyte mass. Burnet proposed that during prenatal development, pluripotent lymphocyte precursors differentiate in the absence of immunogens into a diverse array of individually unique clones of lymphocytes. All of the cells within the clone bear identical immunogen receptors. These events occur while the immune system is still immature. According to Burnet, lymphocytes acquire their specificity prior to immunogen encounter. The capacity in the immune system to recognize and respond to a diverse variety of immunogens is provided by thousands of different lymphocyte clones each with a single, unique specificity. Once the immune system has matured, around the time of birth, an individual possess a full complement of cells with which to recognize immunogens.

Experimentation has confirmed that Burnet's clonal selection theory supports the following concepts: 1) a lymphocyte acquires its specificity prior to immunogen encounter, 2) a lymphocyte's specificity is restricted, 3) lymphocyte activation occurs at the cell surface, and 4) an immunogen selectively expands clones of reactive cells. One would expect that the proportion of an individual's unimmunized lymphocytes that are capable of reacting with any single purified immunogen is quite small. Up to this time, it is possible to account for four characteristics of the immune response: immunity is *acquired* and *remembered* in a *specific* fashion by virtue of clonal selection and expansion. Immunity is *transferred* with antibodies or with lymphocytes. Normal individuals do not mount immune responses against their own cells and molecules. Any theory of im-

munologic responsiveness must accommodate that under normal circumstances the immunologic system is able to distinguish self from nonself (5).

Self Versus Nonself

Burnet proposed that while lymphocyte precursors are proliferating and acquiring diverse specificities during fetal development, lymphocytes of all specificities appear, including cells capable of recognizing cell antigens. During this time, emerging antigen recognizing cells and relevant self-antigens coexist without mutual affect. Then at some defined developmental stage, approximately the third intra-uterine month in humans, cells confronted by relevant antigens (self) are deleted; the remaining antigen reactive cells, which are theoretically capable of recognizing only nonself antigens, persist. Thus, under normal circumstances immunologic mature individuals do not react against self-antigens because self reactive cells have been deleted. Beyond this developmental stage, encounters between nonself antigens and appropriate antigen recognizing cells lead to immunity. In Burnet's mechanism for self tolerance (unresponsiveness towards self), lymphocytes that have specificity for self antigens are called *forbidden clones* and do not participate in adult immunity. Thus, self-antigens are not immunogenic because of the deletion of potentially reactive cells (12).

Hypersensitivity

Hypersensitivity reactions play a very important role in medicine. The *immediate hypersensitivity* reactions are antibody mediated while the *delayed hypersensitivity* is cell mediated. The immediate hypersensitivity reaction can be systemic as seen in anaphylaxis or a local cutaneous reaction with an urticaria response. Immediate hypersensitivity reaction include type 1, type 2 and type 3 reactions (8, 9).

Immediate type hypersensitivity reaction is defined as an inflammatory reaction mediated by antibodies. Immediate type, or antibody mediated hypersensitivities fall into three categories: 1) Anaphylaxis occurs when immunoglobulins of the IgE class interact with antigen thereby causing the release of vasoactive substances which induce an immediate inflammatory response. 2) Cytotoxic hypersensitivity results when antibodies bind to self antigens and destroy the host. 3) Immune-complex hypersensitivity occurs when antigen-antibody complexes are deposited at tissue sites and a subsequent destructive inflammatory response ensues.

Despite its designation, immediate type (antibody mediated) hypersensitivity is not always immediate. The symptoms of anaphylaxis appears dramatically, often within ten minutes following administration of the inciting antigens, but symptoms of cytotoxicity in immune-complex hypersensitivity may take days or weeks to appear.

Type 1. The first type of immediate hypersensitivity reaction is anaphylaxis. The anaphylactic form of hypersensitivity is mediated by the IgE immunoglobulin class. IgE, bound to mast cells or circulating basophils, interacts with the antigen, thereby causing the release of vasoactive substances which induce an immediate inflammatory response. Antigen combines with antibody IgE bound to the cell and initiates release of a vasoactive substance, histamine, on contact with the antigen. An antigen that produces an immediate inflammatory reaction within five minutes of administration in an allergic individual is termed an allergen. The symptoms of anaphylaxis vary with the route by which the allergen is administered. Cutaneously injected allergens produce urticaria (hives) which consist of a pale, edematous area called a wheal surrounded by a red area called a flare. Intravenous administration of allergen can induce systemic anaphylaxis characterized by vasomotor collapse and shock. Both cutaneous and systemic anaphylaxis are caused by the binding of allergens to immunoglobulins of the IgE class. The granules of basophils and mast cells are known to contain histamine, serine esterase and heparin. Histamine, the dominant vasoactive amine in anaphylaxis causes vasodilatation, smooth muscle contraction, increased vascular permeability, and inhibition of further degranulation. Serine esterase is an enzyme that cleaves proactive serum polypeptides into vasoactive substances. Heparin is an anticoagulant capable of eliciting an inflammatory response. Thus, the substances within the granules of basophils and mast cells that are important in the induction of anaphylaxis are histamine and serine esterase. Examples of this immediate type of hypersensitivity are: allergic rhinitis (hay fever) and allergic asthma.

Type 2. The second type of *cytotoxic hypersensitivity* results when antibodies bind to antigens on membranes and destroy the host's cells. Some drugs have a tendency to attach to red blood cells. The body then identifies these cells as being foreign and an antibody response is stimulated. If there is complement present, agglutination and lysis can occur. The cytotoxic category of immediate type hypersensitivity results when antibodies bind to surface antigens located on host's cells. Antibody mediated

cell destruction can occur in several different ways. Antibody binding to cells may activate a cascade of lytic enzymes that normally circulate in the plasma. This system of circulating enzymes is called the complement system. Examples of this type of reaction, are blood transfusion reactions, hemolytic disease of the newborn, and some of the unexplained hemolytic crises that occur while patients undergo intensive drug therapy. Autoimmune hemolytic anemia is probably the best understood example of cytotoxic hypersensitivity. Erythrocyte destruction results from irreversible membrane damage caused by activation of the complement pathway. This activation is achieved when antigen on the surface of host erythrocytes are bound by antibodies. Immunologically mediated hemolytic anemia is seen under three circumstances: 1) Exogenous antibodies directed at host red cell antigens, 2) Host antibodies directed at transfused erythrocytes, and 3) Host antibodies directed at antigens of the host's own erythrocytes. In all three instances, red cell lysis is mediated ultimately through the activation of complement.

Type 3. Immune complex hypersensitivity occurs when antigen-antibody complexes are deposited at tissue sites and a subsequent destructive inflammatory response ensues. If the antigen and antibody combine in the presence of complement to damage local blood vessels, the result is an *arthrus phenomenon*. If there is a systemic effect on lungs, blood vessels and kidney in the presence of excess antibody *serum sickness* results.

Type 4. Cell-mediated or delayed type of hypersensitivity reaction is the fourth type. Cell-mediated hypersensitivity occurs when sensitized lymphocytes react to specific antigens by the release of lymphokines, substances released by sensitized lymphocytes when they come in contact with antigens to which they are sensitized. When PPD, or purified protein derivative of the tubercle bacillus, is injected cutaneously into an individual who has or has had tuberculosis, a significant swelling and reddening appears at the injection site within 12 to 24 hours. This inflammation reaches maximal intensity between one and three days after injection and then slowly subsides. When PPD is injected into a normal individual who has never had tuberculosis, no cutaneous reaction is seen. A positive skin reaction to PPD in the first individual is an example of cell-mediated immunity which is also called delayed hypersensitivity. Delayed hypersensitivity is an immunologic process. It is an acquired and transferable capacity which exhibits memory and specificity for the antigen. The inability of a

normal individual to respond to PPD with significant information without prior exposure suggests that the ability to express delayed hypersensitivity to PPD is acquired. Delayed cutaneous hypersensitivity to PPD can be transferred to an unsensitized individual with T lymphocytes but not with serum, from a sensitized donor. Thus, delayed hypersensitivity, a form of cell-mediated immunity, differs from immediate hypersensitivity which is mediated by antibodies. There are at least four mediators of cellular immunity, including transfer factor (TF) lymphocyte transforming activity (LTA), migration inhibition factor (MIF) and lymphotoxins (LT).

An antigen coming in contact with a sensitized lymphocyte for that particular antigen can cause destruction of the antigen directly, or can produce the very active pharmacologic agents called lymphokines. These give rise to a transfer factor, migration inhibition factor, blastogenic transformation, cytotoxic factor, and interferon. In cell-mediated immunity, there is direct contact of sensitized lymphocytes with the antigen as well as an indirect effect on the lymphokines.

The cell-mediated immunity response of oncology patients may be tested by the use of dinitrochlorobenzene (DNCB). This is carried out by painting dilute solutions of DNCB on three places on the arm. The DNCB acts as a hapten, binding the skin protein to form an immunogen. It is then processed through the macrophage, and having become a processed antigen, it meets with the lymphocytes to sensitize lymphocytes. In approximately two weeks, the patient is challenged again with the dinitrochlorobenzene. If a good delayed hypersensitivity response is identified, that is, erythema and induration greater than 5 mm, it is assumed that patients can mount an immune response against their tumors. There has been high clinical correlation using this test. If the test is negative the patients are usually immunoincompetent and the prognosis is poor as they cannot fight off their tumors. A great number of patients with positive tests show at least an initial response to therapy but the test is not particularly correlated with long term survival.

Immunodeficiency Disorders

Immunodeficiency disorders are classified as defects of the stem cell, T cell or B cell. More than 30 phenotypic patterns have been identified in human beings (15). Among the primary immunodeficiency diseases are hereditary ataxia telangiectasis, Wis-

kott-Aldrich syndrome, selective IgA deficiency disorders and some cases of chronic mucocutaneous candidiasis.

Whenever nature goes wrong, an opportunity to study basic mechanisms is usually brought forth. The Bruton type of agammaglobulinemia (13) consists of repeated bouts of infection that can be controlled by the use of antibiotics but reoccur. In doing electrophoretic patterns on the serum of these patients, Bruton noted that they were lacking in gamma globulin. These patients have a defect of B cell humoral mechanism that make them unable to respond to a challenge with a vaccine by producing immunoglobulins. The deficiency can be controlled by injection of gamma globulin at repeated intervals.

Another syndrome is the Di George syndrome in which there is a deficiency of the thymus and some of these patients are unable to develop a delayed hypersensitivity reaction, cannot fight viruses, tumors or parasitic disease. However, if they are challenged with a vaccine, they can mount an immunoglobulin response, and some patients with the Di George syndrome have the ability to fight off bacterial infections. Recently, they have been treated successfully with embryonic thymus transplants.

The third type of immunodeficiency disorders is the Swiss type agammaglobulinemia. This is a stem cell type of immunodeficiency in which both B and T cells are deficient. These patients usually die in the first two years of life, but recently it has been possible to give some of them successful bone marrow transplants, which restore their B and T cell immunity.

In summary, if the thymus is removed from the new born animal but the gut associated lymphoid tissue is not disturbed, the animal cannot reject foreign tissue grafts or fight a virus but does have normal resistance to bacterial infection. Conversely, if the gut associated lymphoid tissue is destroyed, and thymus kept intact, the animal cannot resist bacterial infection but can reject foreign tissue grafts as well as virus infections.

The clinician today is being offered ways of treating serious immunodeficiency diseases that until a few years ago were impossible to treat. It is now feasible first to determine just what is lacking in the patient's immunologic system and then to compensate for that lack. Thus, a number of patients with Bruton's agammaglobulinemia have been aided with gamma globulin. Bone marrow transplants have given an immunologic system to some victims

of combined immunodeficiency disease, and thymus transplants have helped certain patients with the Di George syndrome (24).

Autoimmunity

Autoimmunity and immunodeficiency are very closely associated. An imbalance in autoimmunity occurs as a consequence of genetic, viral and environmental mechanisms acting singly or in combination. The pathogenesis for autoimmunity may be summarized by saying that there is an excess of helper T cell activity or deficiency of suppressor T cell activity. The overstimulation of the helper T cell can lead to the production of autoantibodies. If this overstimulation continues, as shown experimentally in the New Zealand mouse, a lymphoma or neoplastic process can develop, demonstrating the gradation from autoimmunity all the way to a neoplastic process (16).

The use of very potent drugs, infection, and a neoplastic process often set up a clinical autoimmune disorder which is of a temporary nature as opposed to an autoimmune disease. This has been seen among ovarian cancer patients, who are often very difficult to crossmatch before giving transfusions. It was originally assumed that these women had developed strange antibodies to previous transfusions and had antigens present on the surface of white cells and platelet cells, but in certain instances patients with this problem had had no previous transfusions. The troublesome antibody can be temporarily suppressed by the use of cortisone, and a crossmatch can then be carried out. During the time that they are treated, these patients feel a heightened sense of well being similar to patients who are treated with cortisone who have autoimmune disease.

It has been stated that the immune system distinguishes between self and nonself and responds only to foreign antigens. However, under certain circumstances, the immune system does respond to self-antigens and autoimmune disease results. When the binding of antibody to self or autoantigens results in the destruction of the host's cells, this type of immediate hypersensitivity is termed cytotoxic. Goodpasture's syndrome is a progressive renal disease characterized by the gradual destruction of renal glomeruli with loss of function. Patients with Goodpasture syndrome can be shown to make antibodies against the basement membrane of the renal glomerulus. As a consequence, serum complement is fixed and an inflammatory reaction

ensues which disrupts the integrity of the basement membrane allowing macromolecules in the plasma to be lost in the urine. Goodpasture syndrome is an example of an autoimmune disease.

Complement

Pfeifer noted that the serum of immunized guinea pigs destroyed cholera vibrio organisms in vitro whereas the serum of unimmunized animals did not. He observed further that the capacity of the immunized serum to lyse the micro organisms, which was lost from the serum when the serum was heated to 56 degrees for 30 minutes, could be restored by the addition of unheated and unimmunized guinea pig serum. He concluded that lysis requires at least two different components: one that was produced by immunization (antibody) and the other, that was heat labile and was normally present even without immunization. Since the heat labile components seem to complement the protective function of the antibody, it was termed *complement*. Two serum components responsible for the lysis of cholera vibrio organisms are antibody and complement (21).

Complement is actually a system composed of eleven distinct, naturally occurring serum proteins. There are two pathways for complement activation, the classical complement pathway and the alternate or properdin pathway (23).

The complement components are numbered C 1 through C 9. Activation of the complement pathways begins with the activation of C 1 and proceeds in an orderly (but not numeric) sequence. Immunoglobulins vary in their ability to fix complement. IgM binds C 1 very avidly whereas IgA, IgE and IgD do not bind C 1 at all. The IgG₁, IgG₂, and IgG₃ subclasses fix C 1 but not as efficiently as does IgM. The IgG₄ subclass does not bind C 1. Those immunoglobulins that do fix complement do so only after first binding antigen. The ability of various immunoglobulins to fix complement requires that they bind antigen and then bind C 1 (17).

After an antibody attaches to a target cell, complement also attaches to it through a process C 2 and C 4 enters into the classical pathway and the keystone of this system is complement 3 (C 3). From here on C 5, 6, 7, 8, and 9 combine to lyse the cell or target system. The alternate or properdin pathway is stimulated by polysaccharides, endotoxin, and zymosan which act as the initiating factor. From complement C 3 the alternate pathway and the classical complement pathway are similar.

Immune Complexes

With increasing frequency, reports indicate that a wide variety of chronically disabling diseases are thought to be caused by circulating soluble immune complexes. These include the collagen diseases, that is, vascular diseases, rheumatoid arthritis, glomerular nephritis and possibly chronic inflammatory bowel disease. The antigen-antibody complexes are deposited in the blood vessels of affected organs and initiate damage by activation of the complement system eliciting an inflammatory response. Intensive research using plasmaphoresis as a modality of therapy in the immune complex disease state is in progress (14, 26, 29).

The identification and interest in circulating soluble immune complexes has resulted from the recent introduction of methods that have been developed for detection of these circulating immune complexes and their presence has been now confirmed in a variety of diseases. However, mere detection does not mean that they cause the disease state. Work is now under way to identify what is cause and what is effect. Recently it has been reported that raised levels are found in pregnancy which fall after delivery, suggesting that certain types of complex may modulate the immune response in an advantageous manner, thus possibly preventing maternal rejection of that mosaic of foreign antigens, the fetus. Facilitation or enhancement of malignant tumor growth may also be due to certain types of immune complexes (22).

Immune complexes are being identified with increasing frequency in the field of cancer. Indirect evidence of several tests suggest that immunologic processes accompany many malignancies. Work in progress indicates that immunologic reactions are often occurring in patients with cancer and this observation has been confirmed from frequent demonstrations of antigen-antibody complexes in the plasma of patients and with evidence of the complement system having been activated. The presence of such immune complexes has been demonstrated by a number of different tests, including C1q-deviation test and competition with aggregated gamma globulin receptors on cells of the Raji lymphoblastic cell line. Work at Lenox Hill Hospital has been going on directed to a dissociation of immune complexes. The dissociation of antigen-antibody complexes had originally been effected in the lab at Lenox Hill Hospital by the use of low pH solutions. This method is widely used at the present time and has been successfully applied to our experimental system, however, it presents several in-

herent disadvantages. The low pH levels necessary to effect dissociation of complexes (pH 2.8 to 3), and their subsequent neutralization represents relatively harsh conditions that may result in significant levels of irreversible denaturation of some IgG molecules, rendering them unfit for use in certain procedures such as complement fixation. However, even such extremes of pH had been found to be ineffective in the separation of antigen from antibody and complexes recovered from serum of animals bearing experimental tumors. While low pH is known to dissociate most immune complexes, those with high binding affinities between their antibody and antigen components may be difficult to separate. Our research team has now turned with success to the use of high molarity urea solutions for the dissociation of immune complexes and subsequent recovery of active antibody and antigen. Eight molar urea solutions are highly effective chaotropic agents yielding almost complete dissociation of immune complexes. The denaturation of antibodies produced by eight molar urea is reversible and the removal of the urea by dialysis allows the quantitative recovery of antibody and antigen whose physical properties and immunologic reactivities are unaltered. In addition, since urea is a nonionic, it does not interfere with the separation of dissociated antibody from antigen by ion exchange chromatography. Chromatography of serum proteins or immune complexes on an ion exchange resins yields pure IgG, as ascertained by agarose gel electrophoresis, with subsequent elution of other protein by high salt solutions. This agent is currently in use in our laboratory and is providing us with clean and highly reproducible antibody preparations with substantial tumor related reactivity.

Summary

Immunology is a relatively new discipline of medicine. However, the concept of immunity is an ancient one and properly termed the study of resistance to infection. Immunology cuts across all disciplines. It is interesting that it is a continuum in that at one end the reproductive immunologists are trying to produce tolerance so that the fetus will not be rejected and at the other end the oncologists are trying to produce sensitization so that foreign tissue will be rejected. Modern immunology started when Jenner discovered that inoculation with cowpox cross protected man against smallpox. There have been a great number of scientists from all disciplines that have contributed to the field of immunology. The modern era was considered to

have started when Gross immunized mice by intradermal injection of tumor cells. These mice had been inbred. Having been immunized they were able to reject a subcutaneous transplant of the same tumor while nonimmunized animals were unable to do this. However, the rebirth of tumor immunology as an active field of research may be said to have occurred in 1957 when Prehn and Main observed that mice immunized against syngeneic methylcholanthrene (MCA)-induced fibrosarcomas by inoculation of a living sarcoma tissue, followed by surgical removal of the growing tumor, were resistant to subsequent grafts of the same tumor. It established that tumor cells had substances on the surface that were different from the normal tissue. These were designated as tumor antigens.

It is with some pride that the author notes that many of the early leaders of modern immunology were either trained or taught at Columbia College of Physicians and Surgeons. Dr. Saul B. Gusberg who is being honored by this symposium is steeped in the tradition of this academic environment. He started his spectacular rise to fame at this school.

The following are a few of the scientists from Columbia College of Physicians and Surgeons who contributed to immunology research in its embryonic phase. These scientists include Philip H. Hiss, Frederick Parker Gay, Hans Zinsser, Alphonse Raymond Dochez and Michael Heidelberger. Currently this tradition is carried on by a new group of scientists. These include Dr. Pernis, Elliot F. Osserman, James Halper, Carol Victor, Paolo Tonda, Lotte Kuhn, Dr. Reemstma, Dr. Weber, Carl R. Feind, and Dr. Chess. This is only a small list of those that are making contributions in the field of immunology at Columbia University College of Physicians and Surgeons.

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Connective Tissue Disease in Pregnancy

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Abstract

The diffuse connective tissue diseases, rheumatoid arthritis, systemic lupus erythematosus, polyarteritis nodosa, scleroderma and dermatomyositis and their variants are considered by many to be autoimmune diseases. Since these diseases (particularly rheumatoid arthritis and systemic lupus) are, to a large extent, diseases that occur in females of childbearing age, it is certainly of interest in the immunology of pregnancy to consider the effect of pregnancy on these diseases and of these diseases on pregnancy. In the discussion it will be pointed out that the only two diseases for which there is adequate information are rheumatoid arthritis and systemic lupus. Whereas rheumatoid arthritis often improves during pregnancy, only to flare up afterwards, systemic lupus more commonly exacerbates during pregnancy. The reasons are unclear but certain hypotheses will be presented.

Introduction

The connective tissue diseases that include rheumatoid arthritis, systemic lupus erythematosus, scleroderma, dermatomyositis and polyarteritis are considered by many to be diseases in which autoimmunity plays a major role, particularly in their pathogenesis. The evidence for these is best for rheumatoid arthritis, systemic lupus and polyarteritis and far less for dermatomyositis and scleroderma. Yet, because of overlapping clinical features and some histopathologic similarities, these diseases are often regarded as being in the same category. Certainly full definition of their status and interrelationship depends on better knowledge of the etiologic factors.

Since many of these diseases occur in young women, that is, women of childbearing age, it is relevant in a symposium on reproductive immunology to assess the effect of these diseases on pregnancy and the effect of pregnancy on these diseases. One of the more remarkable points is what little solid information is available. It is my plan to review the various illnesses briefly and discuss their relationship.

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Rheumatoid Arthritis

Rheumatoid arthritis is defined as a disease of the synovial tissues in which there is proliferation and inflammation of the synovium with a tendency toward invasion of subchondral bone, articular cartilage and surrounding structures leading to arthritis and the subsequent destructive arthritic process (1). Associated with this are often systemic manifestations such as subcutaneous nodules, fever, skin rashes, eye problems and, on occasion, vasculitis. The etiology of rheumatoid arthritis is unknown, although there is some speculation as to possible infectious etiology. However, the pathogenesis of rheumatoid arthritis may well be related to an immune complex reaction going on within a synovial space (2). The sequence proposed is that there is immune complex formation within the joint with activation of the complement cascade which releases inflammatory peptides that cause migration of the inflammatory cells through the synovial space. In the process of phagocytizing, the immune complexes release various lysosomal enzymes including proteases and collagenases. These enzymes can therefore cause local damage (3). Much of this hypothesis is based on the finding of immune complexes in the joint, low complement in the synovial fluid, and evidence of complement activation as well as the presence of lysosomal enzymes within the synovial fluid (4). One of the peak incidences of

rheumatoid arthritis is in females of the childbearing age, though there are other peaks, particularly in the postmenopausal female and in the elderly (5).

The disease course is characterized by spontaneous exacerbations and remissions, although in a certain percentage of patients, it is an inexorably progressive one, leading to joint destruction and crippling. Rheumatoid arthritis is considered to be one of the major crippers of young people (6).

Treatment of the disease is empiric involving either suppression with antiinflammatory agents and supportive physiotherapeutic measures or else the use of agents that have the capacity of inducing a remission. There is evidence that agents such as gold (7), Penicillamine (8), Imuran (9) and Cyclophosphamide (9) may indeed stop the ongoing rheumatoid process. The mechanism by which they do so is unclear.

Early in the study of rheumatoid arthritis, it was recognized that pregnancy would frequently induce remission in rheumatoid arthritis (10). Why remissions occur during pregnancy is still not clear, but there is some speculation that the remission is related to the increased concentration of blood cortisol. This observation led to the use of cortisone for the treatment of rheumatoid arthritis (11). This alone cannot explain the remission that occurs in pregnancy. There have been patients who improved during pregnancy, whose cortisol levels were not increased. Other patients with active rheumatoid arthritis who did have increased cortisol levels did not remit. Furthermore, though plasma cortisol level returns to normal within 48 hours after delivery, the remission that occurs usually lasts about six weeks after delivery (12). Other possibilities for the mechanisms of remission in rheumatoid arthritis in pregnancy have been suggested and there has been some enthusiasm for the use of placental extracts, cord serum or blood and urine fractions in rheumatoid arthritis. No consistent improvement has been found (13).

Unfortunately, there is no solid data that explains remissions of rheumatoid arthritis during pregnancy. The incidence of improvement of rheumatoid arthritis in pregnancy is approximately 75%. This number is gleaned both from the literature (14) and personal experience. Improvement seems to begin in the first trimester and seems to continue in the second and third trimester. In about 90% of the patients, however, symptoms recur within two months after delivery (14).

In summary, both from review of the literature and from personal experience, the majority of pa-

tients with rheumatoid arthritis achieve some improvement during pregnancy. However, this improvement is lost within two months after delivery. Although there is a great deal of speculation as to the cause, it is safe to say that the reason for it remains obscure.

Systemic Lupus Erythematosus (SLE)

Another disease in which consideration of pregnancy is important is systemic lupus erythematosus (SLE). SLE affects women primarily in the childbearing age (15). Therefore, pregnancy is a frequent consideration in following these patients. A large number of studies in the literature have evaluated the effect of pregnancy on systemic lupus and the effect of systemic lupus on pregnancy. Although the results of the various studies have been unclear, a general consensus has evolved. Given an individual patient with SLE, the disease can either get better, stay the same, or get worse (16). SLE has manifold manifestations including arthralgias, arthritis, pleuritis, pericarditis, nephritis, nephrosis, thrombocytopenic purpura, anemia, leukopenia, carditis, cerebritis, other neurologic symptoms as well as psychiatric problems (17). The SLE patient may range in severity from a woman who has mild Raynaud's phenomenon and arthralgias, associated with a patchy skin rash, to the patient with a fulminating disease who dies within months of onset. An all-embracing statement about the individual patient with systemic lupus cannot be made.

In personal experience with 23 pregnancies in 22 patients with lupus, the following occurred (Table I). There was one maternal death in a woman who had never had lupus before but developed the symptoms of lupus in the seventh month of pregnancy. It was characterized by fever, arthralgias, proteinuria and skin rash. She was treated for lupus, delivered the baby, and was quite well for 48 hours. Suddenly thereafter, she developed extensive pulmonary hemorrhage with a rapidly progressive course that led, despite aggressive treatment with steroids, cytostatic agents and plasmapheresis, to

TABLE I

*Personal Experience of SLE in Association with Pregnancy
(22 Women, 23 Pregnancies)*

22 Women with 23 Pregnancies	
Mother	Infant
6 SLE flare-up	2 Spontaneous abortion
1 Death	10 Induced abortion
	10 Normal babies
	1 Congenital SLE

her demise within ten days. At postmortem examination, she was found to have severe lupus involvement of all her organs (18).

Two other patients had a flare-up during the third month of their pregnancy. In one patient, it was characterized by hypertension, rapidly progressive renal insufficiency and thrombocytopenia. She improved after a Cesarean section was performed and a healthy baby was delivered.

Another patient had a flare-up characterized by fever, skin rash, and pericarditis. She responded to an increase of her corticosteroids, but had a still birth. Yet another patient developed severe thrombocytopenia, which also improved on corticosteroids.

There were six elective abortions and two therapeutic abortions. Eight children were perfectly normal but one child was born with congenital lupus. The infant had fever, skin rash, anemia and thrombocytopenia which lasted three months, then got better, and never recurred.

Our advice to lupus patients who inquire about the possibility of pregnancy, is that patients with renal disease, particularly when it is active, should not become pregnant. If, on the other hand, the illness has been under control with relatively small amounts of prednisone (10 mg of prednisone/day or less), and they are willing to accept the risks associated with pregnancy, we do not advise against it. Patients on antimalarial therapy of immunosuppressive therapy are advised against getting pregnant because of the possible teratogenic effect of these drugs (19). When the patients do become pregnant, they are followed very closely both clinically and in terms of their serologic abnormalities. With any hint of a flare-up, their corticosteroids are increased.

Some evidence suggests increased fetal wastage during pregnancy. There also seems to be some decrease of fertility in lupus, though this has not been very well documented (21).

Reasons for the flare-up of lupus during pregnancy are totally unknown and there are not even any good speculations.

In summary, systemic lupus can be exacerbated by pregnancy, particularly when there is active renal disease going on. Patients with active renal disease or serious central nervous system disease are advised against getting pregnant. Patients with mild lupus or lupus that is under control with minimal amounts of corticosteroids are not discouraged from becoming pregnant but are followed very closely during their pregnancy.

Dermatomyositis

Dermatomyositis is characterized by inflammatory disease of the muscles and progressive muscle weakness. This is associated histologically with muscle destruction. Often, it is associated with a characteristic skin rash in the periorbital area and over the extensor surfaces of the joints. It is an infrequent, though not rare disease, and affects women slightly more often than men and therefore can occur during the childbearing years (21).

There has been no consistent data about polymyositis and pregnancy in the literature. I have had two patients with polymyositis who have had three pregnancies. All of them were on corticosteroids at the time and they delivered uneventfully. The disease did not seem to affect the pregnancy nor did the pregnancy affect the disease. Except for some scattered cases in the literature, there is no evidence of an important relationship (22). The evidence that dermatomyositis is an autoimmune disease is rather limited. Some patients with this disease do develop antinuclear antibody but its specificity and relevance is not clear (23). There is also evidence of cell mediated myotoxicity (24).

Therefore, there is very sparse data indicating any relationship between dermatomyositis and pregnancy.

Scleroderma

Scleroderma is characterized by vascular phenomena including Raynaud's phenomenon and telangiectasias around the nail beds. There is a progressive thickening of the skin associated with fibrosis and articular findings. Various internal organ involvement include interstitial pulmonary fibrosis, loss of contractility of various portions of the GI tract, myocardopathy and rapidly progressive renal disease (25). The etiology and pathogenesis are totally unknown, but some speculate that it may begin with a disease of the small vasculature (26). The best evidence that it may be an autoimmune disease is that many patients with lupus have antinuclear antibody with a speckled pattern (27).

Patients with graft vs. host disease will sometimes develop lesions similar to that of scleroderma (28). This sometimes also occurs in the animal model of graft vs. host disease (29). The literature is very unclear about the effect of pregnancy on scleroderma, though there have been some isolated case reports of pregnancy exacerbating renal scleroderma with an eclampsia type picture (30, 31).

I saw one patient whose scleroderma initially began during the second trimester of pregnancy and became rapidly progressive after delivery. She remains with a tightly hindbound skin with some esophageal involvement. Another patient with mild scleroderma became pregnant and the disease seemed to have no effect on the pregnancy nor did the pregnancy have any effect on the disease.

Polyarteritis Nodosa

Polyarteritis nodosa is an infrequent disease and its association with pregnancy is rare. I have yet to see a patient with polyarteritis nodosa become pregnant nor have I seen vasculitis complicating the course of pregnancy.

Summary

In summary, the two connective tissue diseases in which pregnancy seems to play a role are rheumatoid arthritis, wherein the general tendency is toward improvement during pregnancy and exacerbation after delivery, and lupus in which the trend can go in either direction. However, some evidence exists that pregnancy can gravely exacerbate lupus, particularly if there is pre-existing renal disease. The mechanisms for both are totally unknown.

In the other connective tissue diseases, there is not enough evidence in any way to assess the influence of one upon the other.

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The Immunologic Concept of EPH-gestosis

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Abstract

EPH-gestosis represents a disease syndrome that is defined by its occurrence in association with pregnancy and by its patient population. Although the difference between "true" and "superimposed" EPH-gestosis remains poorly defined clinically, a clearer definition seems feasible if epidemiologic data, which have been developed in long-term follow-up studies, are utilized. These circumstances permit the identification of a well-defined, young, primiparous population that is afflicted during pregnancy with a disease syndrome of unknown etiology.

Among many suggested theories, the "immunologic concept" of EPH-gestosis has attracted increasing attention over the recent past. Assuming an immunologic etiology for this disease syndrome, EPH-gestosis is understood to represent a partial or total malfunction or maladaptation of the immune system of the mother and/or the fetus. According to this concept, EPH-gestosis is considered to represent partial rejection of the fetal allograft.

In the present review, evidence for an immunologic etiology of EPH-gestosis is summarized. It may be concluded that, in spite of a large body of available information, no definitive proof for an immunologic etiology of EPH-gestosis has yet been presented. Only the further clarification of the immunologic processes involved in the tolerance of normal pregnancy will allow the detection in those mechanisms of abnormalities associated with EPH-gestosis. Until then, EPH-gestosis will remain a condition of unknown etiology. The "immunologic concept," similar to other concepts, needs further clarification until the "concept" can become an "etiology."

Definition of Disease

Edema-proteinuria-hypertension gestosis (EPH-gestosis) is a disease that has eluded clear definition ever since it was first described. This fact is documented most clearly by the many names the condition has been called, even in the recent literature. Many of these names, such as "toxemia of pregnancy," preeclampsia/eclampsia syndrome, and most recently EPH-gestosis, are descriptive of assumed etiologies, signs, and symptoms. However, they confirm that this remains a still largely unknown, and therefore largely hypothetical, disease complex, defined only in a very nebulous fashion.

While most disease states are defined by signs, symptoms, laboratory data, and morphologic findings, EPH-gestosis is defined largely by a patient profile that is in turn defined by the long-term follow-up of patients who had hypertension during

pregnancy (1). Table I summarizes the different profiles of patients with "true" EPH-gestosis and those with "superimposed" disease. However, it cannot be overemphasized that such a differentiation in a clinical situation is purely arbitrary, and that one must rely instead on statistical data accumulated by an exemplary investigator such as Chesley (1). Altcheck was subsequently able to correlate statistical data, at least in part, with morphologic findings in some organs (2).

After it was recognized that hypertension in pregnancy represents at least two major entities, the respective etiologies required investigation. It soon became apparent that in the superimposed group a large number of underlying disease states could lead to hypertension during pregnancy. The etiology of true EPH-gestosis has, however, remained enigmatic and has given rise to a wide variety of different theories, which are listed in Table II.

It is the purpose of this presentation to review the immunologic concept of EPH-gestosis, which has attracted increasing attention in recent years. According to this concept, EPH-gestosis represents a subacute rejection phenomenon of the fetal allograft. The acceptance of this concept would be of

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TABLE I
The Profile of "True" and "Superimposed" EPH-gestosis

	"True" EPH-gestosis	"Superimposed" EPH-gestosis
<i>Predisposition</i>	young primigravidas low socioeconomic groups black patients diabetic patients multiple gestations other conditions associated with "hyper-placentalosis" hydatidiform mole positive family history for hypertension in association with pregnancy	older multiparous female history of essential hypertension history of renal disease family history of essential hypertension obesity low socioeconomic groups black patients
<i>Symptoms</i> (arranged in time sequence of occurrence)	late third trimester Mild sudden excessive weight gain edema develops hypertension develops proteinuria develops Moderate-severe substernal, epigastric pain apprehension, headache, visual disturbances, and nausea hyperreflexia—clonus oliguria Most severe DIC, placental abruption Convulsion	pregnancy and 1st trimester blood pressure frequently elevated no mid-trimester blood pressure dip no associated edema* no associated proteinuria* (or mild in relation to pressure) frequently earlier than third trimester onset frequently more fulminant in blood pressure elevation
<i>Association</i>		Highly associated with IUGR four times the perinatal loss as in the "true" form

*Assuming the absence of underlying renal disease, which may cause either

major importance not only for further research on the biological phenomenon of tolerance, but would also have major implications for the management of EPH-gestosis in the daily routine of a clinical setting. It should be noted, however, that acceptance of the immunologic concept does not necessarily indicate the rejection of any other particular theory on the etiology of EPH-gestosis (Table II). Many of these theories, particularly those descriptive only of biochemical observations, may in fact represent target tissue reactions that were induced by immunologic mechanisms.

Evidence Supporting the Immunologic Concept

The tendency in modern medicine to ascribe all phenomena of unknown etiology to immunologic processes was applied to EPH-gestosis as early as the turn of the century (12, 13). Experimental proof was lacking until mid-century, when Seegal and Loeb ably demonstrated that certain placental antigens cross-react with certain kidney antigens in

TABLE II
Theories on the Etiology of EPH-gestosis (References)

Toxin related (sic! "toxemia")
Primary coagulation defect (3)
Uterine ischemia (4)
Placental damage (5)
Decrease in progesterone (5)
Abnormally sensitive pressor response to angiotensin (6-8)
Prostaglandin defect (6, 9, 10)
Disequilibrium in control of proteolysis (11)
"The immunologic concept"

the rat model (14). The experimentally produced glomerulonephritis, and its similarities with EPH-gestosis, were taken as evidence for an immunologic concept of the latter. Since then it has become apparent that any immune complex state may lead to the deposition of immune complexes within the glomerulus with the same results (15). The morphologic differences, as well as different clinical manifestations, have been well defined (2). However, increasingly sophisticated immunologic investigations of EPH-gestosis have since been performed, and evidence has accumulated indicating a possible

immunologic etiology of this syndrome. It should be recognized, however, that all of the up-to-date evidence remains circumstantial, and final proof is still lacking.

General Evidence

This subheading could read circumstantial instead of general, because the evidence is based on widely accepted immunologic theory that has not yet been proven to be applicable to pregnancy. The profile of the patient with EPH-gestosis represents this type of evidence. Those afflicted are usually young and primigravid women. Previous pregnancy, even of short duration, is known to protect against the development of the syndrome. Extrapolating from other situations in which an antigen is only unsatisfactorily tolerated, it might be assumed that either desensitization or tolerance occurs in the normal female, with the result that EPH-gestosis does not occur. As an explanation for the obvious absence of these protective mechanisms in the young and the primigravid female, one has to assume that desensitization and/or induction of tolerance did not occur either because of age or in the absence of previous pregnancies.

If the former is the decisive determinant, then was the activation purely age dependent, and eventually genetically determined, or rather exogenously induced? Genetic considerations will be discussed under a separate heading. Assuming an exogenous factor, sperm would represent the ideal desensitizing device, because it carries paternal HLA antigens. Desensitization toward paternal antigens could therefore occur prior to pregnancy and depend on the amount of exposure, which would in turn be clearly age dependent. This line of thought was carefully investigated by Marti and Hermann, who demonstrated that exposure to paternal sperm was inversely related to the occurrence of EPH-gestosis (16). But, recognizing the difficulty of any study based on the quantification of exposure to sperm, conclusions cannot be drawn at this time. However, even if pregestational desensitization represents only part of the protective mechanism, a further search for supporting evidence seems to be indicated.

That previous pregnancy is protective against EPH-gestosis has been established beyond all reasonable doubt. However, a clear statistical differentiation between the effect of age versus the effect of gravity remains to be established. Mechanisms which may possibly add to the protective effects of gestation will be discussed later in this chapter; but, some additional supportive evidence for the impor-

tance of age and gravity in protection from EPH-gestosis is worth noting here.

If desensitization and induction of tolerance were dependent on paternal antigen, a certain specificity of the resulting tolerance could be expected. In other words, with the introduction of a new antigenic component, such as a new sex partner, one would expect a return to the levels present prior to desensitization and induction of tolerance in the incidence of EPH-gestosis. Need found evidence supporting such a concept (17), and it has in turn assumed increasing importance in the clinical evaluation of the multiparous patient with hypertensive disease of pregnancy.

The transplant model cannot be omitted in borrowing from immunologic models other than pregnancy. As is true in patients with kidney transplants, whose transplant survival has been shown to improve in recent years if they are transfused prior to transplantation, EPH-gestosis decreases significantly in previously transfused primigravidas (18).

Finally, the puzzling decline in the incidence of EPH-gestosis in most developed countries could be attributable to any of the mechanisms noted earlier. Sexual exposure, and probably the number of blood transfusions, increase concurrently with economic development. All of these factors, in concert with better nutrition, known to have a profound influence on the immune system, could explain the very significant changes in obstetrical praxis in response to the decline in EPH-gestosis in recent years.

Immunogenetic Evidence

The existence of a genetic factor in the etiology of EPH-gestosis has been suggested since Chesley reported an increased incidence of the syndrome in daughters of women who had been afflicted with it themselves (19). Similarly, sisters of women with EPH-gestosis were reported as having a higher incidence than did normotensive control patients (20). The models that were suggested at that time for a genetic theory of inheritance owed much to that of rhesusimmunization. This mechanism, newly discovered at the time, suggested a dominant gene for an antigen that was responsible for incompatibility between mother and fetus (21, 22). The familial occurrence of EPH-gestosis (19, 20), however, refuted such a model. Extreme incompatibility between mother and fetus was still assumed to be the decisive factor, and a large number of studies seemed to confirm this. They included studies on the consanguinity of study populations (23, 24) and the association of EPH-gestosis with a male off-

spring (25, 26), among others. However, a similar number of investigations led to diametrically opposed conclusions (27–29). Other studies on maternal HLA incompatibility yielded inconclusive results.

The extensive review of the problem in recent years (32–35) has best been summarized by an editorial in the *Lancet* (36): Evidence at the present time indicates that the genetic control of severe EPH-gestosis may be dependent on simple mendelian inheritance of a single recessive gene, which may be determined by the genotype of either the mother or the fetus. A final discrimination between the fetal and maternal genotype hypothesis, assuming an immune-response gene, might be rendered feasible by a more detailed examination of family histories (37). It has been suggested that a search for genetic markers (like histocompatibility markers) associated with EPH-gestosis may prove to be of equal importance (36). Thus, extensive further work is needed to determine in greater detail the immuno-genetic background of EPH-gestosis.

Maternal Immune Changes

As the immunologic concept of EPH-gestosis has evolved, increasing numbers of investigators have attempted to demonstrate quantitative changes in the functioning of the maternal immune system in association with EPH-gestosis. Most of these investigations developed as knowledge about normal pregnancy accumulated, and in most of them an attempt was made to differentiate between the findings in normal and abnormal (EPH-gestosis) pregnancy. It soon became apparent that the definition of normal pregnancy was not as clear as had been supposed and that, consequently, the differences between normal and abnormal pregnancy were not as clear. A precise immunologic laboratory definition of EPH-gestosis is still lacking.

As immunologic laboratory techniques have become more sophisticated in general, so have their applications to normal and abnormal pregnancy. Under this subheading we will describe some of the attempts that have been made to define the immunologic profile of EPH-gestosis.

Immunopathology of EPH-gestosis

Since the earliest descriptions of this syndrome, there have been many attempts to identify a specific change in placental immunopathology in association with EPH-gestosis. No such change has yet been reported. The presence of fetus or uterus does not seem to be a prerequisite for the development

of EPH-gestosis, since EPH-gestosis has been reported in association with ectopic pregnancy (38). However, the presence of placental tissue does seem to represent such a prerequisite, and its amount, indicated by placental size, has been correlated with the occurrence of the disease. Thus, EPH-gestosis is closely associated with late gestation, when placental mass is the largest, with multiple gestation, and with molar pregnancy. However, it is not associated with conditions such as hydramnios that produce only uterine distention without an increase in placental mass (39). Most recent studies on placental clearance rates have indicated that placental mass is strikingly greater weeks prior to the onset of clinically evident EPH-gestosis (a time at which placental mass may actually be decreased), than in control patients who do not eventually develop the syndrome of EPH-gestosis (40). These and similar observations have led to the concept of hyperplacentosis, which will be discussed in more detail later.

Some histologic studies have indicated the presence of abnormalities in blood vessels within the myometrium of the uterus. These abnormalities include fibroid necrosis and inflammatory infiltrates in vessel walls, and are called acute atherosclerosis of the vessels (41, 42). The similarity between these lesions and those observed in rejected kidney transplant specimens was taken as further evidence for equating the products of conception with an allograft situation.

Kitzmillier et al pursued this concept by examining decidual vessels for the deposition of various immunoglobulins, complement, and alpha-2-macroglobulin. They observed immunologic specific fluorescence for IgM, C-3, and occasionally IgA, IgG, and alpha-2-macroglobulin in association with EPH-gestosis. However, later investigation revealed similar findings in patients with chronic hypertension and diabetes mellitus, thus invalidating the hopes expressed earlier that these findings were specific to EPH-gestosis (34, 43). Similar findings were reported by a number of other investigators. In our laboratory studies, no specific staining pattern was detectable in either normal or abnormal pregnancies when immunoglobulins and complement were investigated (44).

The immunopathology of the kidney in association with EPH-gestosis has been more clearly delineated. Altchek described the electron microscopy of renal biopsy specimens in patients with EPH-gestosis for the first time in 1961 (2). Later investigations at our as well as other institutions clearly illuminated the renal lesion of EPH-gestosis (45–50). It is best described as a swelling of endothelial

cell (capillary wall) cytoplasm of the glomerulus, a deposit within the cytoplasm and between cytoplasm and basement membrane, and an increase in intercapillary cells. Outside the glomerulus, a juxtaglomerular hyperplasia and a lesion of the loop of Henle were described. Immunofluorescent studies consistently showed fibrin-like material in glomeruli. However, no immunoglobulin or complement deposits were identified. The findings in this respect were remarkably similar to those we observed in placentas (44). Glomerular capillary thrombosis may be found at autopsy in association with many cases of eclamptogenic EPH-gestosis, and has been erroneously interpreted as part of the toxemia lesion. Since then, it has been recognized that it simply represents a complicating terminal reaction (51). Because of maternal complications and associated terminal events, hemorrhagic lesions may be found at autopsy in almost all organs. The most frequently involved organs are the kidneys, the liver, and the pituitary and adrenal glands (51).

Immunoglobulins and Complement

Since immunoglobulin and complement determinations were technically feasible much earlier than more sophisticated procedures (such as the determination of lymphocyte subpopulations), a large number of investigations attempting to correlate the clinical syndrome of EPH-gestosis with immunoglobulin and/or complement levels appeared in the literature. Table III represents a random sample, by no means complete, of such studies and their results. These results have varied widely, depending not only on the laboratory method applied, the investigator, and the timing of the study, but also on variations in study populations, and especially the accuracy of clinical diag-

nosis. In summary, no clear picture as to immunoglobulin and complement levels in association with EPH-gestosis exists at this time. Reported declines in one or both seem rather to be caused by the protein loss that is possibly associated with either true EPH-gestosis or underlying renal disease. Therefore, neither immunoglobulin nor complement evaluations seem to be of any clinical value in the evaluation of EPH-gestosis.

It was noted earlier that immunoglobulins and complement may also be interpreted as deposits. The most typical renal lesion for toxemia was reported to contain fibrin-like material, but no deposits of immunoglobulin or complement could be demonstrated (49). However, later investigations led to different results. Petrucco and coworkers were able to demonstrate a significant correlation between the severity of EPH-gestosis and the density and pattern of IgM and IgG deposits. In addition, complement was found consistently in afferent and efferent arterioles, and in the glomeruli in the most severe cases as well (61). With these observations as a basis, it was suggested that EPH-gestosis may represent an immune-complex state, resulting in the deposition of immune complexes within the renal filtering organ, and thus producing glomerulonephritis, as is seen in association with many other immune-complex diseases (15).

Immune Complexes

To define EPH-gestosis as an immune-complex state or an immune-complex disease, it will be necessary to define these terms. An immune-complex state is defined as a physiologic state in which immune complexes are detected in the circulation by means of a number of standard techniques presently available. In an immune-complex state, the

TABLE III
Immunoglobulin and Complement Levels in Association with EPH-gestosis

Reference	Immunoglobulins				Complement			Comments
	IgG	IgM	IgA	IgD	Total	C-3	C-4	
52					S			
53	L							patients without proteinuria; the diagnosis must be questionable.
54	L	S	S					
55	L	S	S	L				
56		S						
57					S	S		
58					S			
59					L	H	L	only the change in C-3 reached significance.
60	L	L				S		attributes lower Ig-levels to loss in urine

L = lower

S = same

H = higher than controls

presence of immune complexes does not indicate disease. The presence of detectable immune complexes in an immune-complex disease, on the other hand, does indicate a disease state, and may in fact be caused by the presence of immune complexes.

The presence of immune complexes in normal and abnormal pregnancy has been discussed elsewhere (15). The relevance of the subject to the present chapter lies in the recognition that immune complexes may significantly influence the immune system, in both suppressive and activating capacities (15). The definition of pregnancy as an immune-complex state, a question which is still hotly contested, is essential for the definition of EPH-gestosis as an immune-complex disease. At the present time, only a small number of investigators have attempted to clarify the crucial question of the presence of immune complexes in normal and abnormal pregnancy. Among the published studies pertaining to circulating soluble immune complexes, only six investigated sera from patients with EPH-gestosis. A summary of these studies is presented in Table IV. It is evident that the presence of immune complexes in association with EPH-gestosis is still undetermined. Further studies and a clarification of the current detection techniques are urgently called for (15).

If EPH-gestosis is defined as an immune-complex disease, the presence of immune complexes could explain a variety of mechanisms that could in turn explain a number of clinical manifestations the EPH-gestosis syndrome is known to include. The implications of such a concept are demonstrated in Figure 1.

In brief, immune complexes have a known ability to affect the immune response. This effect may be on either humeral or cellular immune response, and

CONCEPTS ON THE INDUCTION OF EPH-GESTOSIS BY THE PRESENCE OF IMMUNE COMPLEXES

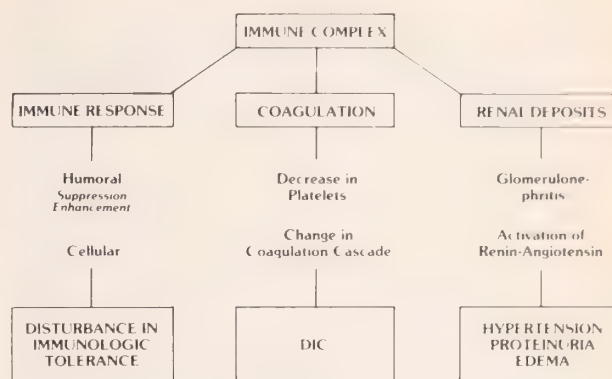


FIG. 1. Concepts on the induction of EPH-gestosis by the presence of immune complexes

may be suppressive or enhancing. Recently, one of us extensively reviewed the details and we have speculated that resulting changes in immune homeostasis may be responsible for disturbances in the immunologic tolerance of the fetal allograft (15). In this sense, EPH-gestosis may represent rejection that is immune-complex induced. The presence of immune complexes in graft rejection has been reported in allograft systems other than pregnancy (68).

Immune complexes also influence coagulation. Platelets and other factors in the coagulation cascade may be adversely affected (11, 69), resulting in diffuse intravascular coagulopathy (DIC). DIC is known to occur in some disease states associated with circulating immune complexes. Some viral diseases such as dengue hemorrhagic fever represent prime examples (68).

Finally, renal deposits may induce a number of events, that may in turn induce hypertensive disease, proteinuria, and proteinuria-caused edema. The renal mechanisms by which these processes may be instigated are poorly understood. However, it seems perfectly reasonable to assume that mechanisms activating the renin-angiotensin axis are induced through immune complexes. Thus, it may be speculated that the loss of reactivity to Angiotensin II, occurring during normal pregnancy and absent in EPH-gestosis pregnancies (8, 79), may only represent the result of immune-complex deposition in the kidney. Stark suggested that the interrelated systems controlling proteolysis may lose their equilibrium during pregnancy, in turn allowing a renin-like enzyme in plasma to produce the effects of EPH-gestosis. He further suggested that the overactivity of this renin-like enzyme may lead to undesirable activities in other physiologic

TABLE IV

*Immune Complexes in Association with EPH-gestosis**

Reference	Number of Cases	Immune Complexes	Comments
62	16	yes	higher levels than normal pregnancies
63	34	no	2/34 were found to be slightly elevated by one of two applied techniques
64	21	yes	72% were positive vs. 11% for normal third trimester pregnancies ($p < 0.0001$)
65	12	yes	9/12 or 10/12, depending on assay
66	10	no	only 2/10 positive
67	14	no	none positive

* Modified from Gleicher and Theofilopoulos (15)

systems, including coagulation and complement (11).

None of the ideas presented here has been confirmed on an experimental basis. It should be recognized that these assumptions are still purely speculative and await confirmation.

Cellular Aspects

As is true with those aspects of maternal immune function in association with EPH-gestosis, only very incomplete data is available on the cellular arm of the immune system in both normal and abnormal pregnancy. A number of the parameters of the cellular aspect of immune function can be investigated with present laboratory techniques (Table V).

Cell Counts

Only very limited information concerning peripheral white blood count alterations in association with normal and abnormal pregnancy has accumulated. Results reported on the composition of the peripheral white blood cell count of pregnancy are relatively few and remain statistically inconclusive. In a very recent study, we were able to demonstrate statistically valid quantitative changes in the peripheral blood of women with normal term pregnancies (71). These included elevations in the absolute number of white cells, segmented neutrophils, band neutrophils, and monocytes. In contrast, a significant decline in the absolute number of peripheral lymphocytes, with a significant number of females showing fewer than 10^6 lymphocytes per milliliter was noted. The percentages of segmented neutrophils and band neutrophils increased significantly as well, while the percentage of peripheral lymphocytes decreased. No significant difference in the percentage of monocytes was observed, however, while the lymphocyte-monocyte ratio de-

creased significantly. These investigations were performed on normal pregnancies. Unfortunately, no similar study has been reported on patients with EPH-gestosis, but two recent studies have attempted to determine lymphocyte populations in EPH-gestosis. Gusdon and associates reported no significant change in total lymphocyte, T cell, or B cell counts associated with EPH-gestosis (72). Birkeland and Kristofferson, in a more sophisticated study, evaluated serial lymphocyte counts, not only in mothers, but also in offspring and fathers, and came to the conclusion that significant changes in maternal, paternal, and fetal cell function may represent the basis for EPH-gestosis (73). However, it may be stated in summary that adequate statistical data on peripheral cell counts is still lacking, quite a surprising lacuna considering the simplicity of such studies.

Cell Activities

Immunocyte function can be evaluated by several *in vivo* and *in vitro* methods. Some that have been used in the immunologic evaluation of EPH-gestosis are listed in Table V. It can be stated that maternal T lymphocyte function, as measured by all of these *in vitro* assays, is generally depressed in patients with EPH-gestosis (74, 75). Once again, however, the data does not seem very convincing and remains to be confirmed. Spontaneous lymphocyte transformation was reported to be the same in normal and afflicted pregnant patients (76), and the reliability of many of the applied assay systems has very reasonably been questioned (75). We described leukocyte migration enhancement as an *in vitro* reaction, specific to both pregnancy and malignancy (77-80), and preliminary results indicate that this enhancement may be diminished in patients with EPH-gestosis. Since we believe that leukocyte migration enhancement depends on a dual mechanism, IgG-induced and possibly suppressor cell dependent, either or both may determine its possible absence in this syndrome.

Cell Interaction

Cell interaction includes the interaction of almost all (white) blood cells in the immune system. Cell interaction in this sense could be equated with cell activity. Red cell function within the complicated and multifaceted immune system is emphasized later by recent observations made in our laboratory which indicate a distinctive function of the red blood cell as an immunocyte, both alone and in interaction with white blood cells (81-83). Similar

TABLE V
Cellular Aspects of Immune Function

<i>Cell Counts</i>
white blood cell differential
lymphocyte subpopulations
<i>Cell Activity</i>
secretion of lymphokines
response to mitogens
lymphocyte transformation
mixed leukocyte reaction
leukocyte migration enhancement
<i>Cell Interaction</i>
autologous red cell adherence
red cell immune adherence

to other very specific immunologic situations, pregnancy was shown to be associated with distinctive changes in red cell immunocyte function, as measured by red cell complement receptor activity.

Fetal Immune Changes

At the present time, it is appropriate to note that at birth the fetal immune system is almost completely developed. Thus, immunologic processes that may represent a part of, or be responsible for, the development of EPH-gestosis may be postulated to be partially or completely dependent on fetal immune functions. Unfortunately, the amount of published data has remained very limited and will permit only partial conclusions.

Immunopathology

Morphologic changes in the fetus itself are no better defined than its immunopathology; and, similar to placental morphology, a classical pathologic or immunopathologic picture of the fetus has not yet been defined in association with EPH-gestosis. Impaired regional blood perfusion of the placenta is considered to be one of the principal features of the syndrome (84). This decreased perfusion may in turn lead to fetal wastage in a number of ways, all of which will result in a specific morphologic picture, related not necessarily to EPH-gestosis but rather to evolving secondary induced pathophysiology. Thus, fetal pathology will present a wide spectrum, depending on maternal clinical manifestations. Evaluating the fetal situation is complicated by the frequently encountered difficulty of differentiating between true and superimposed EPH-gestosis. Thus, so-called preeclampsia of early onset was shown to be associated with growth retardation, low estriol levels, and high fetal wastage, while preeclampsia with late onset was shown to be significantly less malignant (85). It is not clear whether the same disease entity includes both early and late onset or if, on the other hand, two distinct patient groups were thrown into one study. Chesley's data point to the latter interpretation (86).

Immunoglobulins and Complement

Immunoglobulins and complement have been studied extensively in cord blood at various gestational ages and in various obstetric situations (87). However, surprisingly little has been reported on immunoglobulin levels in regard to EPH-gestosis, nor have complement components attracted the attention they deserve.

Cederqvist has recently reported some exciting data indicating that specific changes in fetal immunoglobulin levels may occur in association with EPH-gestosis (88). It remains to be seen if these changes are due to a primary fetal immune function that participates in the generation of the disease syndrome, or if they represent a reaction to an event occurring independently within the maternal immune system. Further work in this field is urgently needed.

Immune Complexes

It is presently unknown if immune complexes cause damage to the fetus in utero, an important question not only in regard to the immune complex controversy over EPH-gestosis, but also as an immunologic question of major theoretical importance (15). Immune complexes may be produced in the fetal circulation, or may have access to the fetal circulation via the placenta. Five theoretical possibilities seem to be feasible:

- *Transplacental transfer of immune complexes from mother to fetus* It is presently unknown whether immune complexes cross the placental barrier. Size represents the determining factor among maternal immunoglobulins. It seems reasonable to assume that the size of immune complexes may be of importance in a similar mode. This possibility indicates the need for a more specific determination of maternal immune complexes, if they are present. This last fact may be of special importance in conditions other than EPH-gestosis, such as associated immune-complex diseases.
- *Separate transfer of antigen antibody and immune-complex formation in the fetal circulation* Antigen may cross the placenta in a variety of forms. Thus, polypeptide aggregates were detectable in animal experiments in the fetal circulation after maternal immunization (89), and viral particles were shown to cross the placenta in many human experiments as well (15). If antigen and antibody cross in a noncomplex state, complex formation may occur in the fetal circulation. This theoretical possibility seems feasible in a situation in which complement absence in maternal circulation does not allow complex formation, while adequate complement in the fetal compartment facilitates the formation of immune complexes.
- *Antigen transfer to the fetus and formation of immune complexes with fetal antibody* A mechanism of this type has been demonstrated in certain animal models (90) and may represent the

mechanism of tissue damage to the fetus in association with TORCH-infections (15).

- *Fetal antigen complexes with transplacentally derived maternal antibody* Rh-disease may be an example of such a reaction.
- *Fetal antigen forms immune complexes with fetal antibody* This situation is present in congenital immune complex disease.

It remains to be determined which of these mechanisms are involved in immunologic processes during normal and abnormal pregnancy, and to what extent. As we noted earlier, no specific damage to the fetus has been reported that could be attributed to the passage or formation of immune complexes in the fetal compartment in association with EPH-gestosis.

Cellular Aspects

The development of fetal white cell activity has been extensively investigated. Unfortunately, only very limited information is available on changes in cellular aspects of fetal blood in association with EPH-gestosis. Thus, T cells appear unchanged and the incidence of B cells is increased in children of women with EPH-gestosis (73). However, no additional significant information is available at present.

The red cell immunocyte function of fetal blood seems to increase in early fetal life (Siegel I, Gleicher N, unpublished material).

Immunologic Interaction

So far we have discussed immunologic changes in the mother and fetus. In our short review of the immunogenetic evidence, we mentioned some evidence indicating that, with increasing consanguinity, a decline in the incidence of EPH-gestosis may be observed (24). We will not attempt to reiterate all the evidence for and against, but one point seems apparent in conjunction with these observations: The immunologic interaction of mother and fetus, and perhaps also the immunologic interaction of mother and father, may play a part in the immunologic explanation of EPH-gestosis.

Early evidence for changes in maternal-fetal immune interaction came from studies with mixed lymphocyte cultures from both mother and fetus. Thus, Gille and associates were able to demonstrate that in normal pregnancies the proportion of fetal cells (84.1 percent) to maternal cells (15.9 percent) was statistically different from that of EPH-gestosis pregnancies, where the fetal proportion reached only 69.6 percent (91). These authors concluded

from their study that the immunotolerant state of normal pregnancy is changed to a state of cellular hyperreactivity in mothers who develop EPH-gestosis.

It is interesting to note that this hyperreactivity seems to be directed solely toward the fetus (indirectly toward paternal antigens), while regular lymphocyte activity, as measured by mitogen stimulation, for example, has been reported as depressed rather than enhanced in EPH-gestosis.

Some evidence supporting the findings of Gille et al comes from histologic studies of para-aortic lymph nodes removed during cesarean section deliveries in normal and EPH-gestosis pregnancies. It was shown that a marked loss of germinal centers may occur in these lymph nodes in normal pregnancy (92), while nodular cellularity increases in EPH-gestosis (93).

In addition, more sophisticated investigations of the immunologic interactions between parents and offspring of normal and abnormal pregnancies were recently reported by Birkeland and Kristofferson (73). By performing serial lymphocyte counts and function tests in parents and offspring during and after EPH-gestosis pregnancies, they defined significant changes associated with this syndrome. When compared to normal pregnancies, EPH-gestosis was associated with all of the changes summarized in Table VI. Contrary to Gille's conclusion, these authors summarized their findings as indicating maternal and paternal hyporesponsiveness together with fetal hyperresponsiveness (73); further investigations are clearly indicated. Birkeland and Kristofferson's findings do deserve special consideration because they also explain the heretofore puzzling finding that during normal pregnancy suppressor cells may be found in fetal, but not maternal, blood (94). A defect in fetal suppressor function could thus be part of the immunologic picture of EPH-gestosis.

Immunologic Mechanisms

If immunologic mechanisms figure prominently in the etiology of EPH-gestosis, then the syndrome must represent the malfunction or maladaptation of immunologic mechanisms that, in normal pregnancies, assure the survival of the fetal allograft within the maternal immune system. Thus, unless the mechanisms involved in the tolerance of fetal products are better understood, neither the etiology nor the mechanisms of EPH-gestosis can be fully understood. In other words, an understanding of the immunologic basis of EPH-gestosis may eventually

TABLE VI

*EPH-gestosis, a State of Parental-fetal Immune Imbalance**

Compared with normal pregnancies EPH-gestosis seems associated with the following changes:
Lower B lymphocytes in the father
Lower T lymphocytes in the mother
Lower B lymphocytes in the mother
Impaired T lymphocyte function in the mother
Low response in mixed lymphocyte culture between mother and father
Increase in B lymphocytes in offsprings

* Modified from Birkeland and Kristofferson (73)

derive not from the immunologic investigation of patients with EPH-gestosis, but rather from outlining the immunologic profile of normal pregnancy. Only when the immunologic profile of normal pregnancy has been established will the abnormality of an EPH-gestosis profile be meaningful. Until then we will have to continue to speak of the immunologic concept of pregnancy, and continue to include it as one among many concepts that are still postulated as causing EPH-gestosis.

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Current Concepts in Hemolytic Disease and Blood Group Incompatibility

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Abstract

The pathogenesis of hemolytic disease of the newborn (HDN) is presented, followed by a discussion of the immune hematologic and obstetric factors determining the disease. The blood group systems and their relationship to HDN are reviewed in conjunction with a discussion of the obstetric management of such cases. Indications for Rh-gamma globulin prophylactic therapy are emphasized in particular.

Introduction

During the past ten years, the routine administration of Rho immune globulin to nonimmunized Rho- (D) negative women, following delivery of an Rho-positive infant or following an early pregnancy termination, to prevent erythroblastosis has represented a great medical advance. Before this method became available, more than 90 percent of clinically severe hemolytic disease of the newborn (HDN) was caused by the interaction of D-positive fetal red blood cells and maternal anti-D alloantibody (1). Rho immune globulin has been very effective in preventing maternal sensitization and, therefore, HDN has become a less common clinical problem. This decreased frequency of anti-D HDN, has focused attention on those unusual cases of significant HDN due to ABO and other fetal-maternal blood group incompatibilities. However, because many patients had already been immunized prior to the availability of Rho immune globulin, total prevention of maternal sensitization has not yet been achieved, and anti-D HDN remains the most important fetal-maternal blood group incompatibility.

Pathogenesis

Hemolytic disease of the newborn (HDN) occurs when fetal red blood cells possess an antigen against

which there are circulating maternal alloantibodies that are capable of crossing the placenta. Various factors are important in determining which of the approximately 400 known red blood cell antigens and their corresponding antibodies can be incriminated in HDN, and to what degree. Table I summarizes these factors.

Certain antigens in the fetal red blood cell are not manifest until after birth. Therefore, little binding of antibody to the red blood cell will occur. A particular antigen may be widely distributed on fetal tissues and not limited to fetal red blood cells (2). In such cases, maternal antibody may attach to these other antigenic sites, leaving less antibody free to interact with the fetal red blood cells.

The nature of the maternal antibody response is also important. Certain antibodies are produced only in response to red blood cell antigenic stimulation. Other, so-called natural, antibodies are produced in response to antigenic stimuli that need not be red blood cell associated. A and B substances, for example, are widely found in nature, and antibodies directed against these antigens are found in individuals in the absence of exposure to A or B red blood cells (2).

Certain antigens are considered to be strong antigens, that is, they are capable of eliciting an antibody response much more consistently than other antigens. Also significant is the dosage of antigen. A blood transfusion containing a red blood cell antigen that the recipient lacks is much more likely to induce sensitization than a pregnancy in which the amount of antigen entering the maternal circulation on fetal red blood cells is significantly smaller (3).

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TABLE I

Immune Hematologic Factors Determining the Pathogenesis of Hemolytic Disease of the Newborn

A. Fetal Factors:
1) Antigen not present in maternal RBCs and present in fetal RBCs
2) Degree of antigenicity of fetal RBCs
3) Distribution of antigen in fetal tissues
B. Maternal Factors:
1) Primary or secondary sensitization
2) Strength and quantity of antigen
3) Magnitude of antibody response
a) Genetic factors
b) Primary or secondary sensitization
c) Strength and quantity of antigen
4) Type of antibody produced: IgM, IgA, or IgG
5) Natural antibody or not

Patients vary in their development of an antibody response to a given stimulus. Nonresponders exist who do not develop antibody despite repeated injections of RBCs possessing a strong antigen (3). The type of antibody produced is also important. Individuals can respond to red blood cell antigens by producing humoral antibody of three immunoglobulin classes, IgA, IgM, and IgG. Only IgG is significant in terms of HDN, since IgA and IgM do not cross the placenta (4).

Anti-D represents the most significant maternal-fetal blood group incompatibility. The D antigen is present in large amounts on the fetal red blood cell membrane. It is not distributed on other fetal tissues, and consequently no competition for maternal anti-D occurs. Anti-D, a very potent antigen, is nearly always red blood cell stimulated, and the antibody produced is largely IgG which can cross the placenta.

The blood group systems and red blood cell antigens will be considered next. HDN may range in severity from merely a positive direct Coombs' test on a cord blood sample (several red blood cell antigens have been discovered in this way), or mild hyperbilirubinemia without anemia, to kernicterus, or the intrauterine death of a hydropic fetus. Awareness of the severity of the condition in utero is essential in order to care appropriately for the patient.

Blood Group Systems

The Rh System

The mechanism of Rh isoimmunization is similar to active immunization with other substances; however, the Rh antigen is present only on the erythrocyte. Therefore, intact red cells, or possibly red

cell debris, must be introduced into the susceptible individual to produce an antibody response. This may occur via the intramuscular, intravenous, or transperitoneal routes. Fortunately, the old practice of giving children parental blood by intramuscular injection as an adjunct in the prevention of various contagious diseases is no longer used. Women are still being immunized to the Rho antigen because of mismatched transfusions and show extremely poor subsequent obstetric performance as a consequence unless they are fortunate enough to bear an Rh-negative offspring. However, the vast majority of sensitized women have been sensitized by some obstetric episode such as delivery of a term infant, abortion, or ectopic pregnancy, but probably not by a hydatidiform mole. The obstetric factors are summarized in Table II.

During placental separation, occurring with any pregnancy episode, breaks may occur in the villi, with subsequent leakage of fetal cells. There are open venous sinuses at the placental site, allowing fetal cells into the maternal circulation. Considering the mechanics involved, the differences in the rate of sensitization will vary with the duration of pregnancy and with the occurrence of specific events.

Levine's suggestion, as early as 1943, that abortion might result in Rh sensitization (5) in Rh-negative women was strengthened by the demonstration of the Rh antigen on fetal megaloblasts as early as the 38th day of gestation. There is thus the possibility of Rh isoimmunization early in the first trimester. Some spontaneous abortions are com-

TABLE II

Obstetric Factors in Determining the Pathogenesis of Hemolytic Disease of the Newborn

A. First Trimester
1) Spontaneous abortion
2) Induced abortion
3) Ectopic pregnancy
B. Second Trimester
1) Spontaneous abortion
2) Induced abortion
a) Invasive: saline, urea, prostaglandin F
b) Noninvasive: prostaglandin E
3) Premature delivery
4) Amniocentesis
C. Third Trimester
1) Premature and term delivery
2) Amniocentesis
3) Antepartum bleeding: placenta previa, abruptio placentae
4) Operative delivery
5) Manual removal of placenta
6) External version
7) Toxemia of pregnancy
8) Multiple gestation
9) Stillborn and anemic fetuses

plete, and the placenta probably separates rather rapidly, closing the venous sinuses quite promptly. Partial separation of the placenta occurs in other cases, providing a greater chance for leakage, and possibly a longer period of open venous sinuses. The need for curettage produces an even greater opportunity for infusion of fetal red cells. An abortion induced during the first trimester also involves partial separation of the placenta and thus a prolonged opportunity for fetomaternal transfusion. Furthermore, fully or partially retained placental tissue is not an uncommon complication of either spontaneous or induced second trimester abortion. A 3.1 percent sensitization rate in patients with first trimester abortions has been reported (6).

Most second trimester induced abortions are performed by intraamniotic instillation of hypertonic saline solution, or prostaglandin F. When placental trauma is associated with amniocentesis, there is often a significant fetomaternal hemorrhage. The sensitization rate is three percent (7).

In otherwise uncomplicated premature labor and vaginal delivery, and in uncomplicated term delivery, the volume of fetoplacental blood is large. The number and size of transplacental hemorrhages were investigated and Jennings reported in his studies on transplacental hemorrhage following delivery that fetal red cells were present in one-third of mothers, resulting in a 15 percent sensitization rate among patients with detectable fetal red cells. He further noted that, as the size of the fetal bleed increased, so did the percentage of patients who became sensitized (8). Papageorgiades reported that, of a control group of women with normal pregnancies and deliveries, 11 percent showed significant fetal/maternal bleeds when they were examined by the Kleihauer technique (9). For other situations the percentages were as follows: Forceps delivery, 33.7 percent; cesarean section, 52.5 percent; manual removal of placenta, 40.3 percent; assisted breech extraction (primigravidas), 25 percent; preeclamptic toxemia (moderate or severe), 32.7 percent; and all causes of antepartum hemorrhage, 30 percent (9).

It may be concluded from these facts that the following events, especially during the latter half of pregnancy, are associated with a greater frequency and/or volume of fetal to maternal bleeding: Placenta previa or abruptio placentae, toxemia of pregnancy, external version, twins, abdominal trauma, and amniocentesis. At the time of labor and delivery those patients undergoing traumatic delivery, manual removal of the placenta, or cesarean section, and those delivering twins, an anemic infant, or a

stillborn of undetermined cause are at higher than usual risk of such bleeding.

The ABO System

Most antibody directed against A and B cell antigens is IgM and, therefore, does not cross the placenta. IgG antibody, however, is common in group O persons but distinctly less common in group A or B persons. Therefore, in cases of ABO-HDN, one would expect the mother to be of group O and the neonate to be of group A or B. Rosenfield has shown that 38 of 39 mothers of infants with serologic evidence of HDN due to ABO incompatibility were group O (10). Since IgG anti-A or anti-B may be produced in response to antigens other than those on red blood cells, it is not surprising that antibody may exist in the absence of prior pregnancy or transfusion and therefore first pregnancies may produce infants with ABO-HDN.

However, there is a distinction between laboratory-detected incompatibility and clinical disease. About one in five pregnancies has the potential of developing HDN on the basis of maternal-fetal ABO incompatibility. About one in 60 pregnancies shows laboratory evidence of HDN. However, only about one in 1000 pregnancies, or one in 200 in which mother and fetus are ABO incompatible, results in HDN requiring treatment (11). Why is this so? One reason is the predominance of IgM, especially in A and B mothers. In addition, A and B antigens are found on fetal cells other than red blood cells. Thus, some transferred antibody is "mopped up" and therefore not available for attachment to the fetal red blood cell membrane (9). In addition, fetal/neonatal A and B antigens are not as well developed as those on adult cells. All of these factors may help to explain the usual clinically mild nature of ABO-HDN and the tendency for the direct Coombs' test on cord cells to be only weakly positive or even negative.

Clinical ABO-HDN usually appears as mild hyperbilirubinemia with only mild anemia, or none at all (3). From an obstetric point of view, it can generally be considered a neonatal rather than a fetal condition, especially in reference to the need for obstetric intervention. Intrauterine death due to ABO-HDN is extremely rare. Laboratory tests are available that will select out mothers who are in potential danger of delivering babies with ABO-HDN. However, the presence of high titer IgG anti-A or anti-B does not necessarily indicate clinical disease in an incompatible baby (12, 13). Moreover, ABO-HDN severe enough to require exchange

transfusion in the newborn of one pregnancy, may be followed by the delivery of an infant in a second pregnancy of the same ABO type who does not develop HDN.

Rh System (Other than D)

Antibodies directed against Rh antigens other than D have been implicated in HDN. These other Rh antigens, for example, C, c, E, and e, are strong, although they are less likely than D to elicit an antibody response. The antibody directed against these antigens is usually red cell stimulated, and is usually IgG. Fetal red blood cells manifest these Rh antigens, and although the reported HDN has usually been milder than that associated with anti-D, severe hemolytic disease can occur (14).

In routine screening of pregnant women's sera for incomplete (nonagglutinating) antibodies, Gibblett found anti-D to be the most common antibody, alone or in combination with other antibodies, directed against Rh antigens. Next in frequency were anti-E and anti-c (14).

Kell System

Antibodies directed against several antigens in the Kell system have been implicated in HDN. The most significant antigen is K, the first discovered. Anti-K, the most common red blood cell stimulated antibody outside the ABO and Rh systems, is largely IgG. Hydrops fetalis with intrauterine or neonatal death caused by K isosensitization has been reported (15–17). Anti-K usually develops as the result of a transfusion with K-positive blood. This is true of the irregular antibodies in general and probably reflects a dosage effect (14).

Duffy System

Anti-Fy A is the most important antibody in the Duffy system in terms of HDN. Although it is less common than anti-K, it is similar to anti-K in several ways. It is usually produced in response to red blood cell stimulation, is largely IgG, and has been found in severe cases of HDN including hydrops fetalis (18, 19).

MN System

The antibodies produced against the antigens M and N are usually IgM and, therefore, HDN only rarely involves these antigens. Other antigens in the MN system, however, do elicit IgG antibody response. Anti-S, anti-s, and anti-U have been responsible for severe HDN (20, 21).

Perspectives

The frequency of irregular antibodies (directed against antigens other than A, B, or D) and the likelihood of resultant serious disease in the antepartum period was demonstrated in a study of 12,297 women, using a standard screening cell (1). Specific irregular antibodies were identified in 85 (0.7 percent) patients. Of these antibodies, 30 percent were directed against Rh antigens other than D, and 25 percent of the antibodies were anti-Le (Lewis). Next in frequency was an anti-K (Kell) with 18 percent; 12 percent were directed against MN system antigens; four percent were anti-Fy (Duffy); three percent were anti-Lu (Lutheran); three percent were anti-P; two percent were anti-JK (Kidd); and two percent were anti-I. Severe disease occurred when antibody had been directed against the non-D Rh antigens, the Kell antigen (K), and the Duffy antigen (Fy). Although M and N are generally not clinical problems, anti-S, anti-s, and anti-U can cause severe disease. ABO-HDN is usually mild, as is HDN due to incompatibilities in the Kidd and Lutheran systems. Although anti-Lewis antibodies are relatively common in maternal sera, and anti-I and anti-P are not rare, these antibodies are not significant in terms of HDN.

Obstetric Management

A careful history is essential. Previous pregnancy outcome is important, especially regarding the occurrence of neonatal jaundice. A previous blood transfusion is a possible cause of maternal sensitization. All pregnant patients should have their serum tested against a screening cell to detect the presence of irregular antibodies. If testing with a screening cell is positive, a cell panel should be used to identify the specific antibody involved. In cases in which maternal IgG antibody capable of causing HDN (for example, anti-K) has been identified, it is important to ascertain whether or not the prospective father possesses the specific antigen in question on his red blood cells. Amniocentesis is indicated in cases when maternal IgG antibody is directed against red blood cell antigens that have been associated with severe HDN and are present on the prospective father's red cells; a listing is given in Table III. Amniocentesis should be performed under sonographic control. Intrauterine fetal transfusion is indicated by the severity of the disease, based solely on amniotic fluid criteria no matter which antibody is involved (22). A direct Coomb's test on cord blood cells after all deliveries

TABLE III

Red Cell Antigens and Hemolytic Disease of the Newborn

- A. Strong antigens capable of producing severe disease; requiring amniocentesis:
- 1) Rh system (D, C, c, E, e)
 - 2) Kell system (K)
 - 3) Duffy system (Fy)
 - 4) MN system (S, s, U)
- B. Weak antigens (mild or no disease); not requiring amniocentesis:
- 1) A & B
 - 2) M & N
 - 3) Lu, LU (Lutheran system)
 - 4) Lewis, I, P

should be a routine procedure because it would alert the pediatrician to potential problems that were previously unidentified or unsuspected.

Rh-immune Globulin Prophylaxis

The concept that passive immunization using anti-D gamma globulin could prevent active Rh isoimmunization was proven by Gorman et al (23). Following this initial work, numerous field trials throughout the world demonstrated that the use of anti-D gamma globulin to prevent Rh sensitization was safe and remarkably effective. Occasional reactions at the site of the injection, but only rare generalized reactions, have been reported, even when the material is mistakenly administered to an Rh-positive individual.

Any situation in which Rh-positive red cells are introduced into the circulation of a patient who is Rh and Du negative and not already isoimmunized is sufficient indication for the use of anti-D gamma globulin. In the United States the standard dose is 300 μ g of gamma globulin administered muscularly within 72 hours of the potentially sensitizing event (delivery or abortion, for example). The timing of the administration is arbitrary, and if by chance the anti-D gamma globulin has not been given during the first 72 hours following this event, it would be given later, since it may be protective and will do no harm (6).

The standard dose of 300 μ g of anti-D gamma globulin will neutralize 15 ml of packed cells or 30 ml of whole blood. The incidence of fetal bleeds larger than this is less than one percent. In the case of first trimester abortion, a minidose of 50 μ g is given (6). The following are suggested regimens for specific patients who are known to be Rh and Du negative and who are not isoimmunized; all should receive 300 μ g of anti-D gamma globulin unless noted otherwise (6).

First trimester (Minidose 50 μ g)

- 1) Menstrual extraction. If performed prior to 38 days from the onset of the last menses, anti-D gamma globulin is probably not necessary.
- 2) Spontaneous abortion with or without curettage.
- 3) Induced abortion.
- 4) Ruptured ectopic pregnancy.

Second Trimester (300 μ g)

- 1) Spontaneous abortion.
- 2) Induced abortion.
- 3) Immature and premature delivery. If the baby can be proven conclusively to be Rh and Du negative, no preventive measures are indicated.

Third Trimester (300 μ g)

- 1) Premature delivery.
- 2) Mature delivery. If the baby is Rh and Du negative, no preventive measures are indicated. If the delivery is complicated by any of the previously mentioned factors that increase the risk of a large fetal bleed, it is advisable to determine the size of the bleed by the Kleihauer technique or the FetalDEX test (Ortho Diagnostics).

Antepartum

- 1) Bleeding. Approximately three percent of patients may have antepartum bleeding, about half of which is caused by placenta previa or abruptio placentae. Unless the father's identity is known, and he is known to be Rh and Du negative, all nonsensitized Rh-negative women who have antepartum bleeding should receive anti-D gamma globulin (6). It is also advisable to determine the size of the fetal bleed and administer adequate amounts.
- 2) Amniocentesis. Patients undergoing amniocentesis should have placental localization prior to the procedure. If an anterior placenta is traversed by the amniocentesis needle, the patient should receive preventive therapy unless the father is known to be Rh and Du negative. If the placenta is not anterior and the fluid is bloody, the cells in the fluid should be tested to determine whether they are adult or fetal and a test for fetal cells should be performed on the mother. Preventive measures can then be taken as indicated (24). Another school of thought routinely gives

anti-D gamma globulin to all Rh-negative patients having amniocentesis.

- 3) External version, trauma, toxemia, antepartum demise. At present it is suggested that such patients be checked for fetal bleeding, and that therapy be given only on appropriate indication (24).

Data from various sources indicate that the use of immunoprophylaxis may be very successful. Accumulated statistics indicate a sensitization rate of 0.18 percent in treated patients, compared to 6.91 percent in untreated patients (24). Robertson reports on accumulated data from several countries that show a 0.2 percent sensitization rate for patients treated after the first Rh-positive pregnancy (25).

Pollock et al report that about two percent of their Rh-negative patients developed anti-D antibody while pregnant or within three days following the birth of an Rh-positive baby (6). On the assumption that some of these sensitizations might be preventable, they attempted to choose those patients who should receive antepartum anti-D immune globulin on the basis of fetal red cell tests, but no improvement in the prevention of sensitization was demonstrated by this technique. Subsequently, they treated all of a series of nonsensitized Rh-negative patients during pregnancy with 300 μ g of anti-D gamma globulin at 28 weeks and also within 72 hours postpartum if the baby was Rh positive; this treatment decreased the rate of sensitization to 0.1 percent (24). Further investigation seems necessary before this method of therapy can be advocated generally, as it doubles the cost to all patients in order to benefit fewer than 2 percent. However, some centers are now adopting these techniques routinely.

The following are suggested as possible explanations for the occurrence of isoimmunization at times other than during pregnancy, although the frequency with which each of them occurs is unknown:

- 1) Physician error. Lack of knowledge or failure to administer immunoprophylactic therapy, even when indicated.
- 2) Laboratory error. A mistake can occur or there may be an error in transcribing the blood type onto the laboratory report.
- 3) Nursing service error. There might be failure to carry out a written order or, again, there may be an error in transcribing the correct blood type from the laboratory report to the patient record.

- 4) Expense. When anti-D gamma globulin first became available, it was very expensive, and some patients refused this therapy because of its cost.
- 5) Religion. Those patients who refuse to allow treatment with blood or blood products refuse the administration of anti-D gamma globulin.

One other rare cause of Rh sensitization may explain the condition of the sensitized patient with no previous conceptions and no history of transfusion: In an Rh-negative baby whose mother is Rh-positive, a maternal to fetal bleed could sensitize the fetus in utero. Fetal immunocompetence is presumed (26, 27).

The value of anti-D gamma globulin in preventing isoimmunization from mismatched transfusion has been supported by Pollack et al (28) and by Keith (29). Each recommends 20 μ g of anti-D gamma globulin for each milliliter of Rh-positive blood received.

The value of administering anti-D gamma globulin to patients undergoing sterilization procedures soon after or at delivery has been discussed. Gorman and Freda recommend that it be given because of the risk of failure of tubal sterilization, and in the event of a possible transfusion reaction at some time in the future (30).

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The Maternal Immune System during Pregnancy and Its Age-related Changes in Rodents

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Abstract

The age-related decline in pregnancy functions of laboratory rodents is illustrated with data obtained from the C57BL/6J mouse throughout pregnancy and at parturition. These data are presented as the basis for a theoretical discussion of a potential causal relationship between reproductive decline and impairments in the maternal immune system with age.

Deficient pregnancy functions become manifest in declining mean numbers of live pups at parturition, in increasing resorption rates during pregnancy, and in the loss of the entire conceptus. Mean litter size in previously parous C57BL/6J mice decreased by over 40 percent from 7.8 pups in three-to-seven-month-old mothers to 4.6 pups in eleven-to-twelve-month-old mothers. Nearly all pups from young mothers were born live, whereas 25 percent of those from older mothers were stillborn. The onset of the age-related increase in resorptions occurred at midpregnancy. Total loss of the conceptus was observed in nearly seven percent of older mothers, but was virtually absent in young pregnancies. The gestation period of older animals was significantly lengthened, and this prolongation was associated with a delayed fall in circulating progesterone, a hormonal change believed to be the triggering event for parturition in rodents.

Since impairments in aged pregnancies may be the result not only of altered uterine functions but also of deficits in the aged ovary, that is, in the number and quality of ovulated ova and/or sex steroid secretion, it was of interest to examine uterine responsiveness in aged animals in the absence of those potentially confounding ovarian alterations. Therefore, ovariectomized mice were injected with a sequence of sex steroids corresponding to the levels found during early pregnancy, and a decidualogenic oil stimulus, considered to mimic that of the implanting blastocyst, was instilled intraluminally into the hormone-primed uterus at a time approximating that of implantation. Under these experimental conditions, the decidual response in uteri of older (ten-month-old) virgin mice revealed marked impairments in several biochemical parameters considered important during early rodent pregnancy, including alkaline phosphatase and glycogen synthesis.

Since decidualization in rodents may be associated with special immune properties that facilitate the retention of the embryonic allograft, it is possible that the observed deficits in decidual responsiveness of the C57BL/6J mouse uterus may, at least in part, be related to alterations in uterine immune properties during early pregnancy. Decidual responsiveness under controlled hormonal conditions and with a standardized, exogenously administered, decidualogenic stimulus may therefore constitute a suitable experimental model for the examination of age-related maternal immune changes during early pregnancy.

Although no detailed experimental evidence is available that causally relates pregnancy failure to deficits in the aged maternal immune system, it is likely that such a causal relationship exists. Therefore, changes in the immune system of young mature pregnant rodents, as reported by several laboratories, are reviewed in this paper as the basis for a theoretical discussion of potential links between pregnancy failure and immune deficiencies in aged rodents. Alterations in properties of T and B lymphocytes during specific phases of gestation are summarized, and the significance of pregnancy-specific immunosuppressive maternal serum factors for the accommodation (nonrejection) of the fetal allograft is discussed.

Since the various phases of pregnancy are largely under hormonal control, and since hormonally regulated changes are a prerequisite for normal pregnancy functions, the specific effects of female sex steroids on several immune mechanisms are described. Relevant experiments from several investigators are presented to illustrate the role of female sex steroids in the modification of immune functions during pregnancy.

Finally, specific points are discussed that may be of relevance for the experimental examination of immune deficiencies and their causal relationship to the overall decline of pregnancy functions in aged rodents.

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Introduction

The paradox of pregnancy depends to a large extent on controlled alterations of the maternal immune system. Those alterations contribute to the accommodation of the embryo which, from the immunologic point of view, constitutes an allograft. The mouse conceptus, like that of other mammalian species, is known to bear histocompatibility antigens (1, 2). Yet despite this antigenicity and the apparent maternal immunocompetence, the fetus is not rejected.

In addition to the pregnancy-related changes in the maternal immune system, the region surrounding the embryo appears to exhibit special immune properties that facilitate the coexistence of the antigenically incompatible mother and fetus (3). This region has therefore been described as an immunologic buffer zone (4). For example, data obtained from mice by the immunoperoxidase labelling technique revealed the presence of minor histocompatibility loci (non-H-2 alloantigens) at the 8-cell stage of the dividing egg. The low initial levels of H-2 antigens observed on the early blastocyst disappeared at implantation. During subsequent embryonic development, no labelling occurred on the ectoplacental cone, which surrounds the embryo; in contrast, the embryonic sac exhibited heavy labelling (5). Similarly, cultured cells obtained from the 7.5 day-old mouse ectoplacental cone had no H-2 and, in most strains, no non-H-2 antigens, whereas 13- to 14-day placental cells in monolayer culture were highly reactive for both H-2 and non-H-2 alloantigens (6). Those and other findings suggest the existence of an immunologically privileged zone surrounding the developing embryo.

On the other hand, ample evidence exists for a pregnancy-specific modification of the maternal immune system as a necessary corollary of successful fertilization, implantation, gestation and, possibly, parturition. The present discussion will focus on selected changes in the maternal immune system during murine pregnancy. Based on the well docu-

mented decline in fertility of aged rodents during a time when the animals are still fertile and capable of giving birth to live pups (7), and based on observations of distinctive age-related changes in various components of the immune system (8), the following presentation delineates evidence for a possible causal relationship between alterations of the immune system and impairments in pregnancy functions in aging rodents.

Impaired Pregnancy Functions in Aged Mice

Alterations in Litter Size at Birth and in Reproductive Capacity During Gestation

In polytocous animals, such as rodents, litter size at birth varies as a function of age and previous parity. The number of live and/or stillborn pups in a species giving birth to multiple offspring may therefore serve as a sensitive index for successful pregnancy functions and for experimentally induced or age-related impairments in those functions.

The C57BL/6J mouse, a strain frequently used to investigate pregnancy-related biologic changes and to assess the deficits in those changes during aging (9), has a comparatively long mean life expectancy of 26 months (10). This strain is therefore particularly well suited for research concerned with primary age-related impairments specific to the female reproductive system, since those impairments commence toward the end of the first third of the total life span, when the major pathologic lesions observed during senescence are absent.

Older pregnant C57BL/6J mice exhibit marked alterations in several pregnancy-related biologic parameters. As indicated in Table I, the mean litter size of previously pregnant mice decreased by over 40% from 7.8 pups in 3-7 month-old to 4.6 pups in 11-12 month-old mothers. Nearly all pups from young mothers were born live, whereas 25% of those from older animals were stillborn.

To relate this net decline in litter size observed at birth to specific impairments in biologic control

TABLE I

Changes in litter size and in the percentage of live births as a function of previous parity and maternal age. (Adapted from Ref. 22)

Age (months)	Reproductive State	No. of Mothers	Total No. of Pups at Birth (mean $\pm$ SEM)	Percent Live Pups at Birth (total no. observed)
3-7	first pregnancy	214	6.5 $\pm$ 0.1	98.7 (1395)
3-7	second pregnancy	19	7.8 $\pm$ 0.4 ^a	99.3 (148)
11-12	retired breeders	37	4.6 $\pm$ 0.3 ^{b, c}	74.9 (171)

Significance: (by Student's t-test)

a) significantly different from above group: $P < 0.01$

b) significantly different from above group: $P < 0.001$

c) significantly different from young, first pregnancy group: $P < 0.001$

Implantation in Aging C57BL/6J Mice

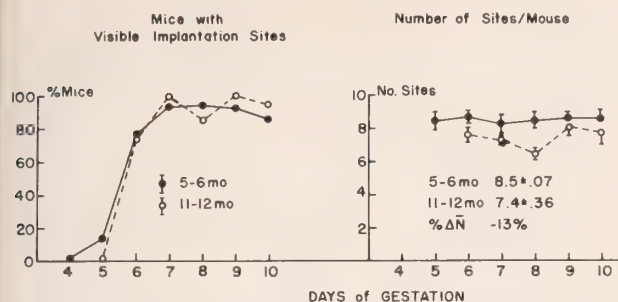


FIG. 1. Percentage of mice showing visible implantation sites (left), and mean numbers of decidual swellings (right) during the first half of pregnancy. The young (5- to 6-month-old) mice were surveyed during their second pregnancy, the older (11- to 12-month-old) mice were retired breeders that had been bred continuously throughout their mature reproductive life span. Day 1 of pregnancy was the first day after continuous exposure to a male on which a vaginal plug was detected. Two C57BL/6J females were placed together with one C3HeB/FeJ breeder male of proven fertility. The number of mice used in this and all other pregnancy experiments appears in Fig. 6.

mechanisms of pregnancy, including those of the immune system, it is useful to examine age-dependent alterations during discrete periods of gestation. Figure 1 illustrates that the number of young and aged mice with implantation sites after impregnation was comparable. On day six of pregnancy (day one was the day when a vaginal plug was found), nearly 75% of the animals of both ages had visible implantation sites which, by day seven, increased to over 90%, without a significant difference in the two age groups. Moreover, there was no evidence that the impregnated aged animals represented in Figure 1 constituted a smaller fraction of the total population than did the young mice (11). Thus, the possibility that the present observations in aged mice applied to a selected subpopulation of especially vigorous individuals was excluded. Furthermore, Figure 1 suggests that implantation occurred at a comparable time in both age groups, since the majority of animals showed decidual swellings on day six, and none on day five.

Taken together, these observations, combined with those of other investigators (12-15), suggest the absence of major deficiencies in the sequence of ovulation, fertilization, implantation and early embryonic development in pregnant aged mice. However, such a conclusion must be viewed with caution since it is based on gross observations which do not estimate quantitative and qualitative abnormalities in the biologic properties of the aged ova and uterus, and which do not resolve potential alterations in synchrony beyond the relatively course interval of one day. Moreover, a small but consistent reduction

in the mean number of implantation sites, amounting to approximately 13%, was apparent already during the early phase (days six to ten) of pregnancy (Fig. 1). Furthermore, the possibility must be considered that biochemical abnormalities at the early stage of pregnancy may become manifest only at a later time.

The period around mid-pregnancy appears to be especially critical for embryonic survival. Figure 2 illustrates the marked decline in the mean numbers of viable fetuses in aged pregnancies between day 11 and 12, and a continuing, small, but steady decrease thereafter. Figure 3 demonstrates a concomitant rise in mean numbers of resorptions (panel A), a decline in the percentage of mothers without any resorptions (panel B), and, after day 12, a consistent small percentage of aged mothers with a total loss of the conceptions (panel C). There is a suggestion of a similar trend in those parameters in young mothers. To what extent these changes reflect a stress point during midpregnancy, and whether such a putative stress point is related to the shift in luteal support from the pituitary to the uterus, which in mice occurs at this time (16), remains open to experimental investigation. The role of the immune system in facilitating any of those processes is as yet poorly understood.

Biochemical Deficits in the Decidual Response

Experimental studies of age-related deficiencies in any of the numerous biologic functions related to pregnancy, including those of the immune system, must dissect the contribution of the embryo from

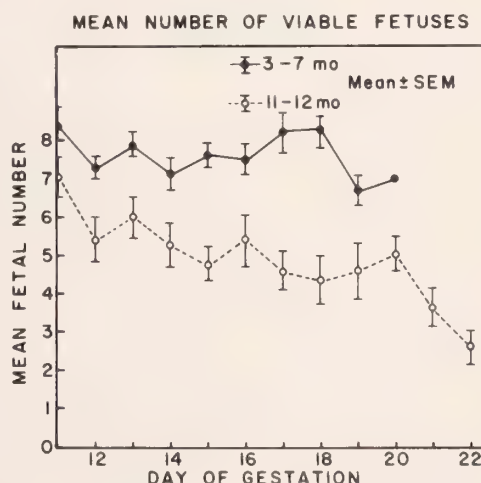


FIG. 2. Mean numbers of viable fetuses in young (3- to 7-month-old) and older (11- to 12-month-old) pregnant C57BL/6J mice. For details, see legend to Fig. 1. Reproduced, with permission, from Ref. 11.

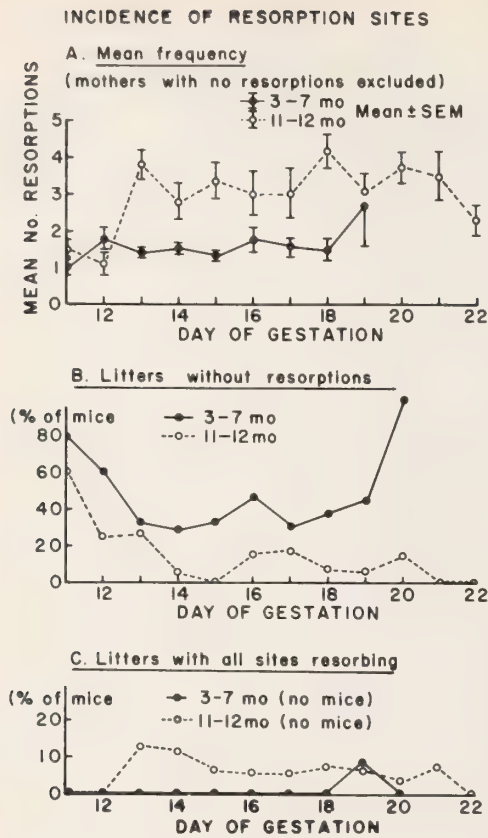


FIG. 3. Incidence of resorptions in young (3- to 7-month-old) and older 11- to 12-month-old) pregnant C57BL/6J mice. For details see legend to Fig. 1. Reproduced, with permission, from Ref. 11.

that of the mother, since both the developing trophoblasts, which represent the aged ovary, and the aging uterus may contribute to the overall reproductive failure. An assessment of the biologic competence of the early pregnant aged mouse uterus has been accomplished by an experimental design which eliminated potential age-related impairments of the ovary in its function as an exocrine and an endocrine gland. Figure 4 illustrates the diminished decidual response in uteri of ovariectomized C57BL/6J mice at an age when initial reproductive impairments become apparent, but when the animals are still fertile. In those experiments, the endocrine function of the ovary was replaced by exogenous administration, after ovariectomy, of an estradiol-progesterone sequence known to mimic hormonal levels during early pregnancy (17), and the decidualizing stimulus normally supplied by the fertilized ovum starting shortly before implantation was replaced by intraluminal instillation of a small amount of arachis oil into both horns of the uterus. Several parameters were examined four days after instillation of the decidualizing oil stimulus. At that

point, maximal decidualization was known to have taken place (13, 14).

The results (Fig. 4) indicate that the response to hormonal priming (column H) was essentially un-

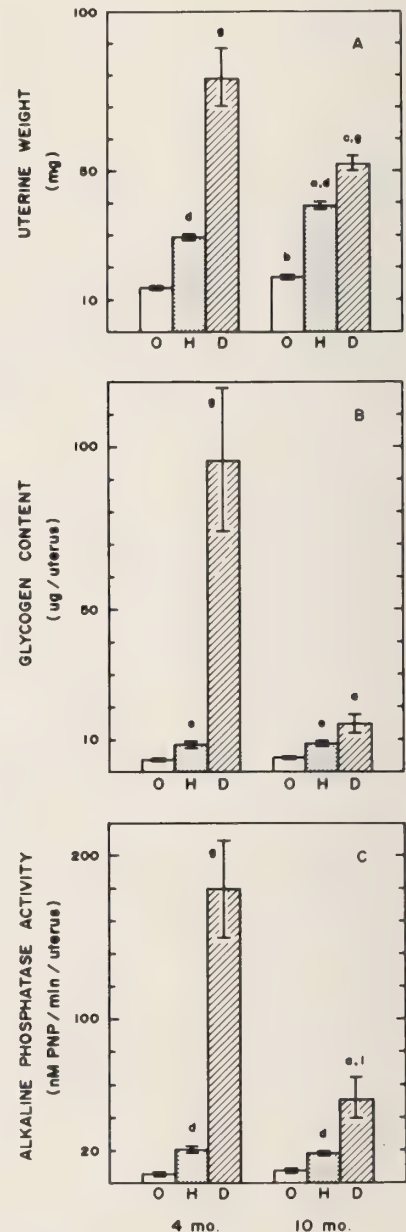


FIG. 4. Changes in uterine wet weight, glycogen content, and alkaline phosphatase activity in ovariectomized: (O) vehicle-injected; (H) hormone-primed; and (D) decidualized uteri of 4- and 10-month-old virgin C57BL/6J mice. Hormones were injected subcutaneously under the dorsal skin as follows: Day 1: ovariectomy; day 3 to 5: 0.1 μ g estradiol in 0.1 ml arachis oil; day 7 to 13: 1.0 mg progesterone in 0.05 ml arachis oil; day 10: 0.025 μ g estradiol in 0.05 ml arachis oil; day 14: collection of tissue. Decidualization was induced 7 to 8 hours after estradiol administration on day 10 with 0.04 ml arachis oil injected into each uterine horn at the utero-tubal junction. Reproduced, with permission, from Ref. 20.

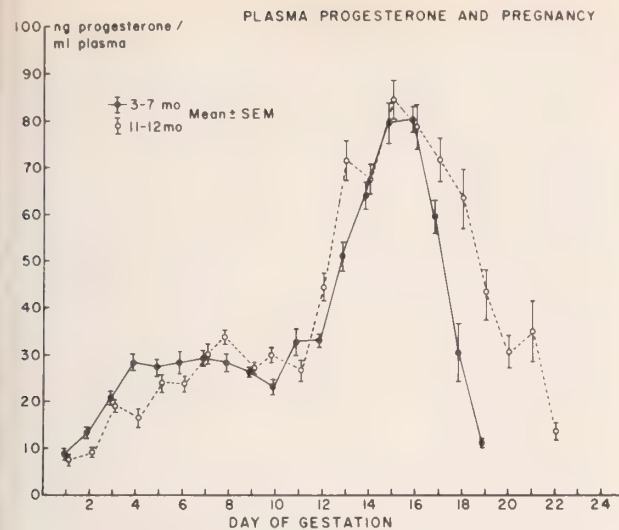


FIG. 5. Plasma progesterone levels throughout pregnancy in young (3- to 7-month-old) and aged (11- to 12-month-old) C57BL/6J mice. For details, see legend to Fig. 1. Reproduced, with permission, from Ref. 11.

impaired in the older mice when compared to the vehicle-injected control group (column O). In marked contrast, the response to the intraluminally instilled decidual oil stimulus was greatly reduced in the older mice (column D). Deficits were observed in uterine wet weight increase, a general index of uterine responsiveness, and in the uterine content of glycogen, a compound implicated in the early postimplantational nutrition of the rapidly proliferating embryo. Deficits were also seen in alkaline phosphatase activity, an enzyme complex involved in the regulation of metabolic energy processes and known to be important during early rodent pregnancy (19). The augmentation in uterine DNA and total protein content (data not shown) was similarly impaired (20).

Taken together, these data suggest deficiencies in biologic responsiveness during early mouse pregnancy that are endogenous to the aged uterus and not secondary to impairments in the aged blastocyst or to ovarian failure. Since the decidual response in rodents has been associated with immunoprotection of the embryo (21), the question may be justifiably posed whether the above described deficits in decidualization are related to alterations in uterine immune properties which, in turn, might contribute to the overall failure of pregnancy functions in aged mothers.

Irregularities in Circulating Progesterone and Estradiol Levels

A considerable body of evidence, derived largely from glandular ablation experiments, selective hor-

monal replacement therapy, and antibody treatment, indicates that progesterone and estradiol are essential hormones in murine pregnancy. Since experimental disturbances in circulating sex steroids result in increased resorption rates or total loss of the conceptus, it was interesting to investigate whether similar deficiencies observed in aged mouse pregnancies were causally related to altered levels in progesterone and/or estradiol. As illustrated in Fig. 5, plasma progesterone levels during early pregnancy of C57BL/6J mice were consistently lower in older mothers, with a significant deficit of 40% on day four, at a period when implantation takes place in mice. Progesterone levels during later pregnancy were comparable in both age groups until day 16, when a rapid decrease took place in younger mothers. This fall in progesterone is considered the triggering event for parturition in rodents. In marked contrast, the decline in circulating progesterone was delayed in older mothers, and this delay was associated with prolonged gestation periods from a mean of 18.6 days in young to 20.8 days in older animals (22).

Deficits in circulating estradiol levels occurred mainly during the preparturitional period, as manifested by reduced rates of increase and diminished absolute concentrations of the hormone in older mothers (Fig. 6).

Pregnancy-related Changes in the Maternal Immune System of Rodents

The modification of the maternal immune system in response to the specific physiologic requirements

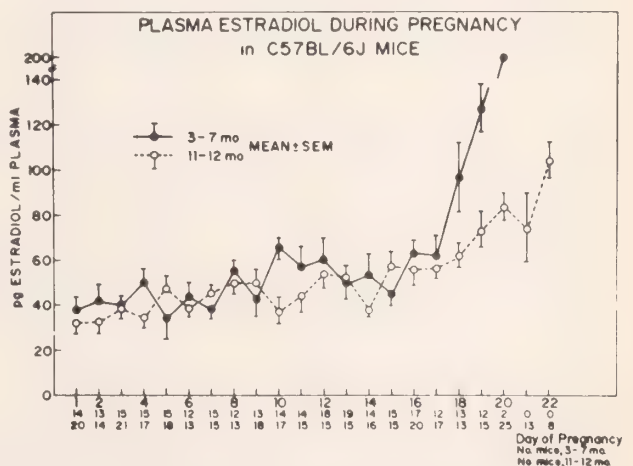


FIG. 6. Plasma estradiol levels throughout pregnancy in young (3- to 7-month-old) and aged (11- to 12-month-old) C57BL/6J mice. The numbers of mice of both age groups used on each day of pregnancy appears at the bottom of the figure. For details, see legend to Fig. 1. Reproduced with permission, from Ref. 70.

of pregnancy is complex. Different components of the system are likely to be modified to a different extent during specific phases of pregnancy. These changes are regulated by a variety of as yet incompletely understood control mechanisms, including the endocrine system (23-28). Table II summarizes a number of pregnancy-specific alterations of the murine immune system during different stages of gestation.

One of the earliest adaptations to pregnancy in female mice is the synthesis of a high molecular weight immunosuppressive serum factor within six hours after fertilization, and considerably before the first division of the fertilized ovum. The site of synthesis of this factor, known as early pregnancy factor, the mechanisms regulating its synthesis and metabolism, and its molecular identity are as yet unknown (29).

The existence of an immunosuppressive factor in mouse pregnancy serum was first proposed from observing that mouse spleen cells, obtained during early pregnancy, were immunosuppressed when subjected to the rosette inhibition test in the presence of antilymphocytic serum against mouse spleen cells, a phenomenon that disappeared during the last third of pregnancy (30). The authors of that study concluded that the immunologic competence

of T lymphocytes was specifically decreased during pregnancy, and that this decrease was not due to the quantitative reduction in the total T-cell population, since the numbers of spontaneously forming rosettes were comparable in lymphocytes of pregnant and nonpregnant animals. Rather, that it was the result of a reduced number of combining sites per cell, so that fewer sites were available for the immunosuppressive action of the antilymphocyte serum. Further work by these investigators established a causal relationship between the pregnancy-related depressed immunocompetence of spleen lymphocytes and the early pregnancy factor (31). They found that mouse pregnancy serum taken at 6 or 24 hours after mating caused the immunosuppression of nonpregnant spleen lymphocytes to levels of immune activity comparable to those observed in the spleen lymphocytes of pregnant animals. The immunosuppressive serum factor was heat stable at 56 C, nondialysable, detectable at a 1:65 dilution of serum (31), and subsequently characterized as having a molecular weight of 180,000 daltons (29).

The immunosuppressive action of the early pregnancy factor, as estimated by the *in vitro* rosette inhibition test, was subsequently clearly established by *in vivo* experiments using the technique of adop-

TABLE II
Pregnancy-related Changes in the Maternal Immune System of Rodents.

Type of Change	Assay	State of Pregnancy	Type of Animal	Source
Appearance of a high molecular weight serum factor associated with immunosuppression of spleen T lymphocytes	rosette inhibition test; gel filtration	6 hours post fertilization to day 12 of pregnancy	Quackenbush mice	29, 31
Immunosuppression of spleen lymphocytes	inhibition of rosette formation between human red blood cells and mouse spleen cells in the presence of antilymphocyte serum against mouse spleen cells	first two thirds of pregnancy, beginning 6 hours post fertilization	Quackenbush mice	30, 31
Rising Clq-binding activity in serum	precipitation by pregnancy serum of radioiodinated purified rat Clq	days 2 to 4	C57BL and CBA mice	36
B cell proliferation in the medulla of iliac lymph nodes	counting of pyroninophilic cells under the light microscope	days 1 to 4	Sprague-Dawley and Wistar rats	37
T cell proliferation in the cortex of iliac and popliteal lymph nodes	counting of pyroninophilic cells under the light microscope	days 1 to 4	Sprague-Dawley and Wistar rats	37
Appearance of a low molecular weight serum factor	rosette inhibition test; gel filtration	following trophoblast formation to parturition	Quackenbush mice	29
Suppressed sheep erythrocyte uptake by the spleen	intravenous injection of ⁵¹ Cr-labelled sheep erythrocytes; splenic uptake measured 15 min later	days 18 to 20	CBA female (crossed with BALB/c male mice)	45
Suppressed macrophage activity	blood clearance of intravenously injected ¹²⁵ I-polyvinyl pyrrolidone	days 12 to 24	A (H-2a) and C57BL (H-2b) mice	47

tive transfer of contact sensitivity to trinitrochlorobenzene by lymphocytes from sensitized mice (32). Lymphocytes were collected from axillary and inguinal nodes of CBA mice four days after sensitization of the animals by skin painting with trinitrochlorobenzene. The sensitized lymphocytes, when injected into isogenic mice, were capable of transferring contact sensitivity, as determined by ear swelling of host mice after painting of the ears with trinitrochlorobenzene. This adoptive transfer of contact sensitivity, however, was suppressed when sensitized lymphocytes, prior to injection into host mice, were incubated with whole pregnant sheep serum, or fractions thereof containing the early pregnancy factor. From this study, it was concluded that the early pregnancy factor suppressed immune properties of lymphocytes *in vivo*, and that its immunosuppressive properties were not species specific, since the sheep early pregnancy factor inhibited the activity of mouse lymphocytes. In addition, it was postulated that the early pregnancy factor served a protective function for the fetus by depressing the immunologic competence primarily of T cells, which are known to mediate both the adoptive transfer of contact sensitivity and the inhibition of rosette formation (32).

Pregnancy-related alterations were also found in the immunocompetence of peripheral blood and splenic lymphocytes of several rat strains. Those cells, under *in vitro* conditions, were fully capable of recognizing paternal alloantigens, yet that capability was suppressed by maternal pregnancy serum (33).

A somewhat contradictory line of results, however, indicates that during certain periods of pregnancy, murine spleen cells appear to be endogenously suppressed by mechanisms not attributable to serum factors. For example, the phytohemagglutinin response of spleen cells from syngeneically as well as allogeneically mated mice was significantly reduced on day 15 in one study (34), and on days 15 and 18 in another (35). It has been proposed that this suppression may not be a primary one, but that serum components not related to the immunosuppressive serum factor described above may complex with phytohemagglutinin and thus bar its accessibility to lymphocytes, or that such compounds may block phytohemagglutinin receptor sites on the lymphocyte membranes (34). Whatever the reason, the effect appears to be restricted to a discreet period of pregnancy between days 15 and 18.

A unique, pregnancy-related change in the maternal immune system involves a marked rise in serum Clq-binding capacity in several strains of

mice (36). This rise is restricted to the early phase of pregnancy. It is most pronounced between days two and four, and is followed by a fall below non-pregnant levels after day seven. The significance of this initial increase in Clq-binding activity, comparable after syngeneic and allogeneic matings, is obscure. Whether it represents an initial and transitory humoral response to fetal antigens or, alternatively, constitutes an early step in a larger entity of immune changes that contribute to the overall nonrejection of the embryo remains open to experimental inquiry.

An immune response during early rat pregnancy has been postulated on the basis of morphologic changes in the iliac and popliteal lymph nodes, where a marked increase in the number of T lymphocytes was demonstrated in the thymus-dependent cortical areas between days one and four of pregnancy (37). In contrast, B-cell proliferation occurred only in the iliac lymph nodes. As with the rise in pregnant rat serum Clq-binding activity, the proliferation of T and B cells did not constitute an immune response to antigens only of a foreign strain, since it occurred in both syngeneic and allogeneic pregnancies, although the increases tended to be somewhat lower after inbred matings. The results of this study were interpreted to indicate that antigens derived from spermatozoa, seminal fluid, and/or embryonic tissues were capable of entering the maternal circulation, since an immune response took place not only in the regional iliac lymph nodes that are accessible to antigens via the regional lymphatic vessels, but also in the distal popliteal nodes (37).

The seemingly contradictory observations that embryos from allogeneic matings could produce a maternal immune response which, paradoxically, did not trigger their own rejection, were further clarified in several studies by K. Hellström and coworkers. These investigators reported (38) that the *in vitro* colony formation of target cells obtained from C3H mouse embryos was significantly reduced in the presence of lymph node cells taken from regional nodes of pregnant BALB/c mice that had been immunized with C3H embryonic tissue. Sera from pregnant BALB/c mice, after mating with C3H but not with BALB/c males, abrogated this inhibitory effect of the specifically immune lymph node cells. This abrogation was more effective after several pregnancies, but it did occur at primiparity. A subsequent report (39) indicated that cells taken from 12 to 15 day-old embryos of isogeneically mated BALB/c mothers were killed by lymph node cells from inbred pregnant mice of the same strain.

Furthermore, lymph node cells from inbred pregnant BALB/c mice had a cytotoxic effect on tumor cells derived from that strain. The precise relationship between the antigens on BALB/c embryonic cells and those on transformed cells, both being recognized by pregnant-mouse lymph node or spleen cells, is not clear. It has been cautioned that transformed-cell antigens may be the result of a virus or of some type of contamination rather than true embryonic antigens (39).

The cytotoxic effect of pregnancy lymph node cells on tumor cells could be blocked by BALB/c pregnancy serum; however, this pregnancy serum blocking activity disappeared after absorption with tumor cells, incubated at 37°C (39). There is evidence that the pregnancy serum blocking factors are associated with immunoglobulins of the IgG class. Immunoabsorbents charged with anti-mouse IgG_{2a} or IgG_{2b}, but not with anti-IgM or anti-IgA, removed the blocking activity, which could be recovered in the eluates (40).

The molecular structure of the pregnancy serum factors that block the cytotoxic effect of pregnancy lymph nodes on tumor cells, and the relationship of those factors to the immunosuppressive serum factors found during normal pregnancy remain to be clearly established. The serum factors regulating immunosuppression during early mouse pregnancy may differ from those during late pregnancy. For example, a low molecular weight serum fraction corresponding to approximately 40,000 daltons, was isolated during later pregnancy (29), in contrast to the 180,000-dalton molecular weight fraction detected in early pregnancy (29). The precise biochemical relationship between those two molecular species remains to be determined.

Among a number of immunoregulatory mechanisms during pregnancy, alpha-fetoprotein, elevated in the maternal circulation (41), has been implicated in the suppression of both cell-mediated and humoral immune responses in mice (42). A selective suppression of antibody formation in mice in response to sheep red blood cells was demonstrated in vivo at mid-pregnancy, and pregnant mouse spleen cells were similarly suppressed in vitro (41). Additionally, antibody production by spleen cells from nonpregnant females was markedly suppressed by pregnant mouse serum; that suppression could be abrogated by selective removal of alpha-fetoprotein by affinity chromatography (41). Also, alpha-fetoprotein was shown to induce specifically the formation of highly active suppressor T cells which were able to inhibit helper T cells, without significantly affecting B cells responding to thymus-independent antigens (43).

In this context, it is of interest that pregnant C3H mice showed a significant decline in allergic reactions after induction of contact allergy at least five weeks prior to the onset of pregnancy by the application of picryl chloride to the shaved abdomen. During the ensuing pregnancy period, the allergic reaction in response to the application of the sensitizing agent to the ear, which was determined by the increase in ear thickness 24 and 48 hours after its application, was greatly suppressed (44). In contrast to this suppression of cellular immunity, the antibody response was elevated during the middle third of pregnancy in C3H and in Swiss albino mice, as measured by the number of plaque forming cells in response to intraperitoneally injected sheep red blood cells (44). This finding opposes the above observation of pregnancy-related selective antibody suppression in mice (41), a contradiction which may in part be due to differences in the strains used in those two studies or, as has been suggested (41), due to differences in methodology.

Early uptake of ⁵¹Cr-labelled sheep erythrocytes after intravenous injection was found to be significantly suppressed in the spleens of 18- to 20-day-pregnant mice (45). This suppression, which indicates reduced phagocytic activity, could be experimentally produced by estrogen injection into non-pregnant mice, and has therefore been ascribed to the rising estrogen levels at the end of pregnancy. Although these findings suggest a functionally significant preparturitional suppression of phagocytic activity in the spleen, this conclusion lacks force because nonpregnant mice rather than animals during earlier stages of gestation were used as controls and, furthermore, the pregnant, estrogen treated animals did not augment their antibody production in response to sheep erythrocytes (45).

The complexity of immunoregulatory mechanisms during pregnancy, some of which restricted to discreet time periods during gestation, is exemplified by macrophage activity in the pregnant mouse. Macrophages are known to regulate humoral and cell-mediated immune responses, as shown in vitro, for example, by the restoration of immunocompetence in macrophage depleted mouse spleen cells after the addition of macrophages (46). Nicklin and Billington (47) evaluated macrophage activity during mouse pregnancy by surveying the blood clearance of radioiodinated polyvinyl pyrrolidone following the intravenous injection of that compound into A (H-2a) or C57BL(H-2b) mice. In allogeneic as well as syngeneic matings, polyvinyl pyrrolidone clearance was significantly reduced between days 12 and 14 of pregnancy, whereas no significant differences were noted between virgin

mice and pregnant females at earlier stages of gestation. The control mechanisms regulating this temporary suppression of macrophage activity during mid-pregnancy, its biologic significance, its age-related alterations, if any, and their consequences for the survival of the conceptus, are at present not known, as is the case with the majority of the above outlined pregnancy-related changes in the maternal immune system of rodents.

The Role of Female Sex Steroids in the Control of Immune Functions and in the Adaptation of the Maternal Immune System to Pregnancy

The sequence of pregnancy, including fertilization, implantation, gestation and finally parturition, is essentially under hormonal control. It is therefore justified to examine to what extent the maternal immune changes summarized in Table II, which presumably constitute a prerequisite for successful pregnancy, are regulated by hormones. Similarly, a causal relationship may reasonably be stipulated between the age-related decline in pregnancy functions, illustrated in Table I and Figures 1 to 3, and a deficiency in circulating hormones and/or target tissue responsiveness to those hormones. As an extension of those considerations, it is tempting to propose the following, albeit speculative, syllogism: that hormones regulate immune changes which are required for successful pregnancy; that age-related hormonal impairments have been found in pregnant rodents; and, thus, that the age-related decline in live births and the increased resorption rates in aged pregnancies may in some fashion be related to deficiencies in hormonally regulated immune functions. Such a proposition, however, awaits thorough experimental examination.

The interrelationships between the endocrine and the immune systems have been under experimental investigation for several decades. A role for female sex steroids in the control of immune functions was first stipulated on the basis of sex differences observed in several immune responses. For example, proliferation of antibody producing spleen cells in response to *E. coli* exposure of mice was significantly greater in females than it was in males (48, 49), a difference that could be abolished by ovariectomy (48), and enhanced by estradiol treatment of mature as well as immature mice (48, 49). Similarly, increases in hemolysin titers in response to sheep red blood cells were greater in females of several strains of mice than they were in males (50), a sex difference which likewise was attributed to

the action of estrogens, since it was augmented by estradiol administration (50).

A series of studies in mice by Nicol, Vernon-Roberts and coworkers gave further support to the role of female sex steroids in the control of immune functions. These investigators demonstrated *in vivo* fluctuations of reticuloendothelial activity during the estrous cycle. Highest phagocytic activity occurred during proestrus and metestrus, as determined by the clearance of intravenously injected carbon (51). Estradiol, estriol and estrone strongly enhanced reticuloendothelial functions, in contrast to progesterone, which stimulated the system only to a minor extent (52), and which did not inhibit the estradiol-induced increase in reticuloendothelial activity (53). The enhanced ability of the reticuloendothelial system to clear carbon after a single injection of estradiol occurred in two phases, with the short-term response within 15 minutes after intravenous injection apparently due to activation, and the 12-hour response due to proliferation of cells in the reticuloendothelial system (54). Enhanced T and B cell activity also occurred in two peaks in four-day cycling mice, as assessed by spleen cell responses to T and B cell mitogens, and by antigenic challenge with sheep red blood cells (55). Mitogenic responses of spleens from BALB/c mice to *in vitro* exposure to concanavalin A, phytohemagglutinin or *E. coli* endotoxin were greatest in cells obtained during proestrus and metestrus, and the mitogenic activity of untreated cells was likewise highest during those periods. The antibody response to immunization with sheep red blood cells, determined *in vitro* by hemolytic plaque formation, was also greatest in proestrous or metestrous mice (55).

Levels of circulating female sex steroids were not determined in the above experiments. Plasma progesterone in rodents shows patterns during the estrous cycle similar to those of the above described immune changes, with one peak at proestrus and another, smaller peak at metestrus (56, 57). The possible biologic significance of this correlation remains to be demonstrated. It has been suggested, but not experimentally validated, that pregnenolone, a progesterone metabolite, may be responsible for the peaks in immune reactivity during the estrous cycle (55).

Estradiol, unlike progesterone, rises in a single peak. This rise begins during diestrus and reaches its highest level at proestrus, as was demonstrated in the C57BL/6J mouse (58). A single estradiol peak, however, does not automatically exclude the possibility of biphasic immune changes regulated by estradiol. As will be recalled, reticuloendothelial

functions showed two peaks of enhanced activity in response to a single estradiol injection, the first apparently caused by activation of the existent cell population, the second caused by proliferation (54).

In contrast to its activating effect on the reticuloendothelial system (51-53), on T and B cells (55), and on antibody responses (55), estradiol appears to have a suppressive effect on natural killer cells, a type of lymphocyte lacking the characteristics of mature T or B cells, which in mice are most prominent in the spleen and in peripheral blood (59). Mice that were chronically exposed to estradiol, released from implanted silastic capsules, showed significantly suppressed natural killer cell activity. This suppression was thymus independent and reversible, could be induced in both male and female, gonadectomized or intact animals, and was attributed to a reduction in cell proliferation of bone marrow precursor cells. A direct effect of estradiol on the thymus was excluded, since that hormone also reduced natural killer cell activity in neonatally thymectomized mice (60).

The thymus appears to be an essential organ for proper functions of the female reproductive system. Athymic female nude mice have incompletely developed ovaries and atrophic uteri (61); their puberty is delayed and the mature females are usually sterile (61, 62). An interaction between progesterone and the thymus is suggested by the presence in thymocytes of 20- α hydroxysteroid dehydrogenase at an activity at least 16 times higher than that in tissues from lung or kidney. The enzyme is likewise present in the spleen of intact C57BL mice, but absent in the athymic nude mouse spleen; its activity was shown to be restricted exclusively to T cells (63).

Evidence has accumulated for an immunosuppressive function of progesterone, likely to be of special importance for the retention of the fetal allograft during pregnancy (25, 64). Exogenous administration of progestins resulted in a marked prolongation of the period of allograft rejection (65) and of the primary humoral antibody response in several species (65, 66), and the resistance to oncogenic viruses was substantially lowered after progesterone injection (64). Skin homografts transplanted into uteri of pregnant or nonpregnant, progesterone-treated rats survived significantly longer than those of nonpregnant control animals (67).

In contrast to its suppressive effect on cellular immunity, progesterone markedly enhanced the production of circulating antibodies in response to bovine serum albumin injected into ovariectomized rabbits (65). The survival of skin grafts, however, could not be prolonged (65). One explanation for

the conflicting results pertaining to the capacity of progesterone to suppress cell mediated immunity is related to the relatively low circulating progesterone concentrations in plasma, when compared to the considerably higher intrauterine concentrations during pregnancy, when progesterone has been implicated in immunosuppression. Siiteri and coworkers (25) demonstrated a significant antiinflammatory, T cell-mediated suppressive effect of progesterone in concentrations approximating those found in the pregnant uterus, in experiments involving the subcutaneous implantation of Syrian hamster skin into rats. Implants wrapped around a progesterone releasing silastic capsule or placed in the immediate vicinity of such a capsule, were retained up to five weeks, in contrast to the complete rejection of those implants, within one week, by nonimplanted animals or by those with progesterone implants at a distant site. Corticosterone, the most active natural glucocorticoid in the rat, and deoxycorticosterone, but not cortisol, had similar effects.

Evidence for the hormonal control of pregnancy-specific alterations in immune functions is at present primarily based on the correlation between changes in circulating sex hormones and concomitant changes in several immune functions which, in nonpregnant conditions, are known to be enhanced or suppressed by sex steroids. In examining such relationships, one must beware of stipulating causality, especially so since antigenic challenge, such as that imparted by the embryonic allograft, in itself may have a primary effect on hormonal changes, and since steroidal and nonsteroidal hormones other than the principal female sex steroids and their metabolites may influence immune functions.

Within that framework of limitations, however there is convincing evidence for interrelationships between female sex steroids and altered immune functions during pregnancy, although the precise biologic meaning of those alterations is frequently not well understood. Some of this evidence is as follows.

Two peaks of phagocytic activity were observed during mouse pregnancy, one at implantation, the other before parturition (51). Since estrogens are strong stimulators of the reticuloendothelial system (52), and since circulating estradiol levels are rising at, or shortly before those periods of enhanced macrophage activity, it is justifiable to stipulate a relationship between the rise in estradiol and the activation of the reticuloendothelial system as, indeed, had been done at a time before precise quantitative measurements of steroid hormones became possible by radioimmunoassay.

By similar considerations one might speculate that other components of the immune system known to be under estrogen control, such as the above discussed natural killer cells (60), the proliferation of antibody producing spleen cells (48, 49) or the altered T and B cell mitogenicity (55), would undergo appropriate modifications in response to peripheral estrogen changes during rodent pregnancy.

The primarily immunosuppressive action of progesterone, like the stimulating activity of estradiol, also appears to be enhanced during pregnancy. Skin homografts, for example, survived significantly longer in uteri of pregnant rats than did those in uteri of nonpregnant animals (67), and skin allografts, implanted subcutaneously, were successfully retained under the influence of high topical progesterone concentrations which approximated those in the pregnant uterus (25), a finding which demonstrates a role for progesterone in the suppression of cell-mediated immune responses during pregnancy and, in addition, touches upon the limitations of conclusions concerning hormonally mediated functions that are based exclusively upon changes in the levels of circulating hormones.

Conclusion

This presentation postulated a relationship between the reproductive decline in aged pregnant mammals, as illustrated by data from the pregnant C57BL/6J mouse, and age-related impairments in the maternal immune system, a biologic system that undergoes characteristic changes in adaptation to pregnancy, as summarized by data obtained from young rodents in several laboratories.

The extent to which deficits in the maternal immune system contribute to the overall decline in murine pregnancy functions is at present poorly understood. Experimental data on correlations between the magnitude of reproductive decline and the degree of failure of immune functions are not available. It is conceivable that aged pregnant mothers exhibiting total loss of the conceptus are less capable of activating pregnancy-related immune changes, such as those summarized in Table II, than are mothers with partial resorptions. Alternatively, partial losses of the conceptus may represent failure of local immune mechanisms restricted to individual fetoplacental units. The primary deficiency in this case may reside in the biologic makeup of the embryo derived from the aged ovum. Thus, the relationship between embryonic loss and chromosomal abnormalities remains to be investigated, and interesting insights are likely to originate

from comparisons of chromosomal patterns of viable versus resorbing embryos within the same conceptus. Correlations of numbers of resorbing embryos with the extent of maternal immune deficiencies may aid in establishing whether embryonic loss is the cause or the effect of impaired maternal immune mechanisms. This consideration is warranted since, in sheep, it was shown that induced embryonic death as well as surgical removal of the embryos resulted in a prompt decline of the early pregnancy factor in maternal serum without any interference with circulating progesterone levels (68).

Another area of fruitful experimental inquiry into age-related impairments of the maternal immune system of rodents pertains to immunologic mechanisms that are specifically restricted to certain phases of pregnancy. For example, during early pregnancy, and specifically on day 4, the day of implantation, circulating progesterone levels of older pregnant C57BL/6J mice were found to be reduced (Fig. 5). Since a number of characteristic adaptations to pregnancy occur at that period in the immune system of young rodents, as illustrated in Table II, and since those adaptations are likely to be at least in part regulated by progesterone, it is of interest to investigate the relationship between reduced progesterone, embryonic loss, and maternal immune deficiencies at that particular phase of pregnancy. Similar considerations apply to the immune changes typical for later stages of pregnancy, and to their age-related alterations.

The exogenously induced rodent decidual response, whose immune components are as yet poorly understood in young as well as in old animals, is a potentially profitable research model for the investigation of age-related deficiencies, since this model is designed to minimize interfering impairments by other components of the maternal brain-pituitary-ovarian axis.

Finally, the well-being of the conceptus after birth is itself a crucial endpoint for the role of the maternal immune system during gestation and for the consequences of age-related malfunctions of that system. Experiments may be designed to investigate the capacity of the immune and other biologic systems in the offspring at various stages of their lifespan, and to correlate deficiencies, if any, with age-related impairments of maternal immune functions during pregnancy.

Acknowledgments

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The Development of the Fetal Immune System

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Abstract

The immune system develops a unique capability of generating practically unlimited immune specificities. This capacity, however, must be restricted since significant immune responses against autologous antigens could induce lethal autoimmune reactions. To solve this problem, the fetus develops tolerance responsive periods during which exposure to many antigens results in tolerance rather than immunity. This eliminates cells of high affinity for autologous antigens. The specificities of the fetal immune system are, however, the result of the adaption of the immune system, not only to autologous but also to the maternal antigens. The placental barrier is incomplete and allows the transfer of both antigens and immunocompetent cells. In this way, the maternal immune responses against the fetus, and fetal immune responses against the mother, may be induced. Fetal immunocompetent specific cells that can eliminate invading maternal cells must, therefore, be allowed to develop. At the same time, specific cells that can damage the maternal host must be suppressed. Thus, immune specificity is affected by an adaptation of the fetal immune system to two hosts and to a delicate equilibrium between the potential protective and injurious effects of the immune response.

Introduction

Philosophers and biologists alike have never ceased to wonder about the development of a human being from a single cell during a gestational period of approximately 40 weeks. At two to three months gestational age, the fetus has already laid the foundation for a complex and sophisticated immune system. The development of the immune system is the result of the same inductive and recognition factors that give rise to other major organs. The immune system must, however, contend with additional unique circumstances.

The present review has to remain somewhat limited in its scope but is directed toward illustrating some of the unique developmental aspects of the immune responses. Most important amongst these may be the observation that the survival of the fetus is dependent on its immunologic adaption to

two hosts, the autologous and the maternal. Similarly to life itself, the immune system seems to represent a very delicate balance between mutually contradictory forces.

Fetal Tolerance

The immune system develops the ability to mount a specific reaction against a practically unlimited number of antigens. Included in the cell population generated by the immune system are immunocompetent cells which are capable of reacting against autologous antigens. Consequently, a special adaptation of the immune system is required to selectively eliminate or suppress cells that are capable of damaging the autologous host. The fetus survives in its maternal host for the gestation period without either rejection by the maternal immune system, or a fetal graft versus host reaction against the mother. Neither of the potential immunologic disasters occurs despite the placental barrier between mother and fetus being permeable, thus allowing for the mutual transfer of cells as well as certain soluble antigens and antibodies (1). Therefore, an additional adaptation of the fetal immune system is required which enables the fetus to survive in its maternal host.

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The adaptation of the fetal immune system to autologous and maternal hosts requires tolerance activities in three different areas:

- 1. The fetal immune system must suppress fetal immune responses against fetal (autologous) antigens.
- 2. It must suppress maternal immune responses against the fetus.
- 3. It must suppress fetal immune responses against the mother.

Mariya (2) and Oldstone et al (3), have demonstrated relatively high suppressor T-cell activity in cord blood. They suggested that the suppressor cells function in the diminishment of maternal immune responses against the fetus. We suggest that a nonspecific fetal circulation T-cell factor is likely to play a simultaneous suppressor role, not only in one tolerance requiring area but in all three areas mentioned above. Further research is needed to elucidate the mutual effects and interrelationships between the triple tolerance requiring areas.

The establishment of fetal tolerance to autologous and maternal antigens requires the elimination or suppression of immunocompetent cells which possess the potential to react against those antigens. The inactivation of such a cell population also inactivates immunocompetent cells which potentially can cross-react with certain pathogens. For example, certain group A hemolytic streptococci exhibit immunologic cross-reactivity with human heart muscle (4). An infection with these bacteria may, therefore, cause autoimmune reactions, damaging heart muscle. On the other hand, induction of tolerance to autologous heart muscle would eliminate immunocompetent cells that could react against the bacteria. Consequently, a balance must be established between the induction of tolerance against autologous and maternal antigens and the possession of immunocompetent cells that must guard the fetus against foreign pathogens or maternal immunocompetent cells.

Cellular Aspects

Hematopoietic Centers

Stem cells that give rise to the immunocompetent cells of the immune system (5) originate in hematopoietic centers at a number of locations that change with developmental stage (6). Table I gives an overview. There is a considerable overlap in the activities of these foci. The liver is the most active site until the middle of fetal life, when activity starts to decrease slowly, until it ceases at birth. The red

TABLE I
*Overview of Hematopoietic Centers which Produce Stem Cells at Different Developmental Stages**

Organ	Developmental Stage
Yolk sac	4th week
Body mesenchyme and blood vessels	4th week
Liver	6th week
Spleen, liver, and lymph nodes	2nd-4th month
Bone marrow	4th month

* Modified from Arey LB: Developmental Anatomy (6)

marrow becomes dominant at five months and eventually serves as the single source from which red blood cells and granulocytes form. The non-granular elements are then mostly supplied by the lymphoid organs.

The Development of the Thymus

Of the essential immune developmental events, many can be illustrated by following the development of the thymus (6, 7). The thymus is evident at about six weeks of gestation. It remains mostly epithelial until populated by lymphocytes at about nine weeks gestational age. The origin of the first thymic lymphocyte is controversial. Early work suggested that lymphocytes differentiated from either thymic epithelial cells or mesenchymal cells that have penetrated into the thymic epithelial anlage (6). Newer studies, however, suggested that most, if not all, lymphocytes appearing in the thymus are cells that have migrated into the thymus from outside hematopoietic centers (6). This migratory cell theory is supported by the observation that, when an animal is transfused with an outside source of thymocytes, very few of the thymocytes settle in the thymus (8). Lymphocytes that enter the thymus continue to differentiate along the T-cell line under the inductive effects of the thymus. Boyse and Abbot (9) were able to demonstrate this inductive effect of the mouse thymus in vitro. They incubated primitive prethymic lymphocytes with thymic extracts. The mouse prethymocytes acquired after a short incubation time in thymic extract typical antigenic T-cell markers.

However, early differentiation towards the T-cell line has occurred before the cells have entered the thymus. This was made evident by not only thymic extracts but also a variety of nonspecific factors that could trigger the differentiation of the prethymic cell. Amongst such nonthymic factors were tissue extracts, cyclic AMP, endotoxin, and synthetic polynucleotides (9). This early thymus independent differentiation towards the T-cell line may account for the cells with certain T-cell character-

istics, such as the ability to transform in response to allogenic lymphocytes (10) or to respond to anti-T-cell serum (11), which are found in the liver before the cells have been exposed to the thymus.

Developmental inductive factors are required for certain immune responses throughout life and may account for the essential role of cellular interactions, such as those between B- and T-cells.

When the thymus becomes populated with lymphocytes, an extraordinary antigen independent growth process is initiated (1, 7, 12). Until the third trimester of pregnancy, the thymus weight/body weight ratio constantly increases. After that, the thymus continues to grow in size at a slower rate until puberty, when a gradual degeneration process sets in.

Autoradiographic studies of the thymus have indicated that the medium and large thymic lymphocytes divide approximately every eight hours (12). Such a rate of division is equal to the rapid growth of lymph node lymphocytes which occurs only in response to intense antigenic stimulation. The life span of the thymic lymphocyte within the thymus is approximately three days (12). Under such a circumstance, the thymus would occupy approximately 10^{12} cubic kilometers in 30–40 days. In fact, the actual weight of the thymus in a two-month-old infant can be weighed in grams. Thus, we can conclude that thymocytes are destroyed at a rate only slightly slower than their production rate. Only a small percentage of thymocytes dies a "natural death" during mitosis. *So, where do all the thymocytes go?* One of us was able to demonstrate that thymocytes adhere to autologous macrophages (13). Furthermore, the thymocytes are often phagocytized and destroyed by the macrophages (Fig. 1). Macrophages from newborn guinea pigs are often more active than macrophages from adult animals (Siegel I, unpublished data). Autologous macrophages may, therefore, through their killing function, account for at least a portion of the disappearing thymocytes.

The eventual size of the thymus is thus the result of an equilibrium between both extraordinary growth factors and extraordinary destructive macrophage forces. These remarkable growth and destruction patterns have been thought to play a major role in the generation of large antibody specificities and the induction of tolerance in several basic theories of immunity (14, 15).

PHA Responsive Cells

Lymphocytes appear in human fetal blood at eight weeks, in the thymus at nine weeks, and in

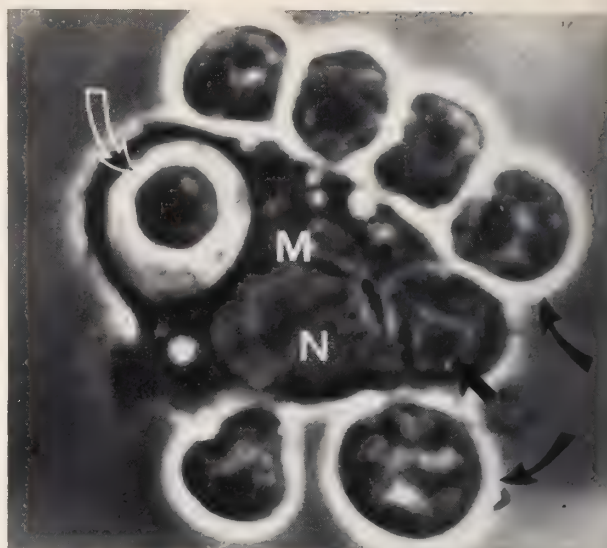


FIG. 1. A guinea pig peritoneal macrophage (M) has phagocytized two autologous thymocytes (black straight arrow and white arrow). One thymocyte (white arrow) has been partly digested by the macrophage. Six thymocytes (curved black arrows) are adhering to the macrophage. The nucleus (N) of the macrophage is easily distinguishable from the thymocytes. The reaction occurred *in vitro* in the presence of 30 percent autologous serum after two minutes incubation at room temperature.

the spleen at fifteen weeks gestational age (16). The sequence of appearance of lymphocytes with T-cell characteristics has been reviewed by Loke (1) and Miller (17). Practically all characteristic T-cell responses have been detected during fetal life. The ability of thymocytes to respond to PHA, a known T-cell mitogen, is evident at ten to 14 weeks of gestation and may be elicited in lymphocytes of the spleen and blood two to four weeks later. Liver and bone marrow cells never develop this ability. A comparison of fetal to adult activities yielded values of PHA response which may be either lower, equal, or higher in either. Another characteristic of fetal lymphocytes is their ability to transform, with no or merely the slightest outside stimulation, into blast cells (18). This observation suggested that these cells had been already stimulated *in vivo*. Such stimulation may have occurred through exposure to maternally derived antigens. We suggest that maternal antibodies against fetal lymphocyte surface antigens represent another likely source of stimulation.

Mixed Lymphocyte Reaction (MLR)

The mixed lymphocyte reaction (MLR) represents the earliest detectable T-cell response. It occurs in liver lymphocytes at about seven and one-half weeks gestational age. Stites et al (10) postulated that this phenomenon plays an important role

in the elimination of maternal lymphocytes which have the potential to act against the fetus. If the fetus contains cells that can respond against maternal antigens, then a complete tolerance to maternal cells—despite early exposure of the fetus to the antigens—has never been induced. Once more this illustrates the balance between tolerance and immunity in association with normal pregnancy.

Lymphotoxin

A deficiency of lymphotoxin production of newborn lymphocytes was reported by Eife et al (19). Lymphotoxin levels of the newborn were about 25 percent of adult levels, although there were no significant differences in blast transformation responses to PHA.

Lymphocyte Red Cell Adherence

In 1970, the adherence of a relatively large proportion of human lymphocytes to sheep red cells was seen for the first time (20–22). Consequently, these lymphocytes were identified as T-cells (23–25). Wybran et al (25) reported that 65 percent of thymus lymphocytes, at 15 to 16 weeks gestation, form rosettes with sheep red cells. In an 11-week-old fetus, only 15 percent of the lymphocytes form rosettes. Hallerach and Komlas (26) observed that the proportion of rosette-forming cells (RFC) in cord blood is inversely related to that found in the mother's blood. Davis and Galant (27) and Smith (28) reported significantly decreased percentages of RFC in cord blood, while Ferguson et al (29), reported no significant difference between normal adults and normal newborn infants. These differences in results may be, in part, due to differences in methodology.

The adherence of human T-cells to sheep red cells does not play a physiologic role because T-cells do not usually encounter sheep red cells. However, a physiologic role of the sheep red cell receptor on T-cells can be assumed by following up on an observation made by Siegel and Sherman (30), who demonstrated that rabbit and rat thymocytes adhere to autologous red cells (Fig. 2). This observation led one of us to the hypothesis that the primary role of the sheep red cell receptor on lymphocytes may lie in a receptor function for autologous red cells (31, 32). In contrast, the adherence of sheep red cells to this receptor is, within such a framework, considered to represent the result of a fortuitous cross reactivity of sheep red cells with the T-cell autologous red cell receptor. This theory led to the prediction that human red cells would adhere

to autologous red cells before the phenomenon had been experimentally established (32). Autologous T-cell erythrocyte adherence has since been demonstrated by a variety of laboratories (33–40).

Only relatively few studies compared fetal autologous red cell thymocyte activities to those of adults. Siegel and Sherman demonstrated in the rabbit that newborn thymocyte-autologous red cell adherence exceed that of adult animals (Table II) (30). Preliminary results in our laboratory have likewise shown that autologous lymphocyte, red cell adherence of human cord blood exceeds that of adult lymphocytes (Siegel I, Gleicher N: unpublished data).

The adherence of thymocytes and T-cells to autologous or homologous red cells represents another manifestation of an antigen independent recognition phenomenon. Thymocytes and T-cells are able to discriminate between red cells and other cell types and between red cells of homologous and heterologous species (30).

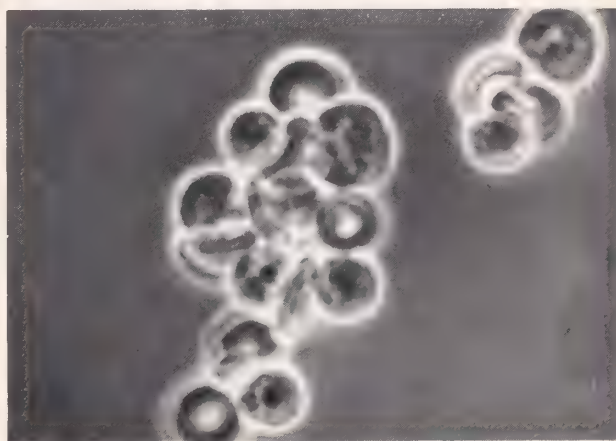


FIG. 2. Rabbit thymocytes and autologous red cells in modified Hanks solution containing 25 percent autologous serum (phase photomicrograph). Mixtures were examined after incubation for 60 minutes at 37° C. From Siegel I, Sherman W: Lymphocyte-autologous red cell interaction (30). With the permission of the publisher.

TABLE II

*Effect of Age of Rabbit upon Rabbit Thymocyte-red cell Adherence**

Source of Thymocytes	Percent of Adhering Thymocytes (Mean $\pm$ S.D. of 5 Duplicate Slides)	
	Red Cells from Newborn	Red Cells from Adult
Newborn	74.9 $\pm$ 7.1	79.1 $\pm$ 7.1
Adult	42.3 $\pm$ 5.2	39.6 $\pm$ 5.0

Cells were suspended in 100 percent modified Hanks solution and observed after sedimentation.

* Modified from Siegel I, Sherman WB: Lymphocyte-autologous red cell interaction. (30) With the permission of the publisher.

The purpose of autologous T-lymphocyte red cell adherence is unknown. We have postulated that it represents a manifestation of immunocyte function of red cells (41). Red cells adhere to antigen-antibody-complement complexes through the red cell immune adherence (RCIA) phenomenon. The red cells then adhere to T-cells, thus presenting the T-cells with antigen. This is illustrated in Figure 3. Our preliminary results indicate that RCIA activity of fetal red cells exceeds RCIA activity of adult red cells (42). Thus, both red cell functions, autologous red cell-T-cell adherence and RCIA, are stronger in the fetus than in the adult.

Humoral Aspects

B-cells

Undifferentiated lymphocytes and partially differentiated B-cells, migrating from hematopoietic centers, are believed to settle in the bursa of Fabricius equivalent of the human fetus, where they complete their differentiation towards the B-cell line. The localization of the bursa equivalent areas in the fetus is uncertain (43). B-cell ontogeny has been reviewed by Loke (1) and Miller (17). A view of recent studies is given by Siegal (44).

Lymphocytes with Ig-bearing receptors are evident in the fetal liver at nine to ten weeks of gestation. At about 14 and one-half weeks of gestation, the proportion of Ig-bearing B-lymphocytes reaches an adult level. Surface Ig classes seem to appear sequentially induced by an antigen independent process. The order of development is Ig-M, Ig-G, and Ig-A bearing lymphocytes. Ig-D bearing

lymphocytes followed Ig-M bearing lymphocytes, but the temporal relationship of the Ig-D bearing lymphocyte to Ig-G or Ig-A bearing lymphocytes is undetermined (17).

Synthesis of Antibody

Although the proportion of fetal Ig-bearing B-lymphocytes is close to adult levels, the synthesis of antibody by the fetus is clearly below adult levels (45). Several studies, utilizing the incorporation of radiolabelled amino acids into immunoglobulins, demonstrated fetal synthesis of Ig-G at low levels. Ig-M was shown at 10.5 weeks, Ig-G at 12 weeks, and Ig-A at 30 weeks (17). Congenital infections are accompanied by a raised cord serum level of immunoglobulins, especially involving the Ig-M and Ig-A classes (46). Cooper et al (47), and Bridges et al (45), concluded that a specific factor is required for the differentiation of lymphocytes with surface Ig-G to antibody synthesizing plasma cells. The fetus and certain agammaglobulinic patients may have a full assortment of Ig-bearing B-cells, but are deficient in a factor that is required for the conversion of these cells to plasma cells.

Complement

The fetus is not only deficient in antibody synthesis but is also deficient in the synthesis of complement components. Synthesis of complement has been detected as early as 5.5 weeks gestational age (17). It thus precedes the synthesis of immunoglobulin. There is little or no placental complement transfer (1, 17). Several reviews of the literature (1, 17) have indicated that, in general, the fetal complement value is 50-75 percent of adult values. This relative complement deficiency augments the general deficiency of the fetal effect on immune function. In collaboration with Dr. Stephen Lubicz and Kenneth Francus, we have detected complement inhibitory factors in cord sera. This factor became evident by observing that the addition of cord blood to mixtures containing adult natural antibodies against sheep red cells prolonged the time required for complement-induced lysis of sheep red cells in the mixture (Table III).

General Comments

While the fetal immune system must adapt to both the autologous fetal host and the maternal host (possibly utilizing similar mechanisms), there is a clear time distinction between these two adaptations. The adaptation to the autologous host has

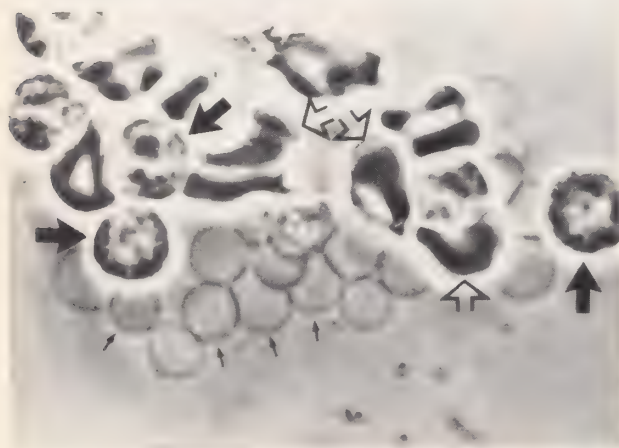


FIG. 3. A mixed aggregate containing rabbit red cells (open arrows) which have adhered to autologous thymocytes (large black arrows), and sheep red cell ghosts (small arrows). The sheep red cells lysed in response to the rabbit antibody and complement, and adhere to the rabbit red cells through the RCIA phenomenon.

TABLE III

*Effect of Cord Serum on Complement Activity**

% Adult Serum	% Cord Serum	Lysis of 0.03% Sheep Red Cells
17%	0%	11½ minutes
17%	8.5%	13½ minutes
17%	17%	15½ minutes
17%	34%	24½ minutes
17%	68%	45 minutes

* Adult serum contained natural antibody against sheep red cells and complement. Mixtures were incubated at room temperature and became transparent when the sheep red cells lysed.

to last a life-time, while the adaptation to the maternal host has to last for the period of gestation only. Such a mechanism would seem beneficial if it could prevent an immune response against maternal antigens without the induction of tolerance to the involved antigens. This is accomplished by a temporary weakness in the effector mechanisms of the immune response which include low levels of antibody and lymphotoxin production, low levels of complement, and the presence of complement inhibitory activity. We postulate that this occurs in part as an adaptation of the fetus to its maternal host. The fetus can generate immunocompetent cells that can react with maternal antigens without causing actual strong reactions against the maternal cells. This, however, renders the fetus and the newborn relatively susceptible to infection and to graft versus host reaction by maternal lymphocytes which have penetrated the fetal immune system (48). Here, again, a balance must be established between contradictory forces suppressing on the one side and enhancing for the immune response on the other. This balance allows the fetus to synthesize some antibody and complement but leaves the newborn susceptible to certain infections (49).

Conclusions

The major components of the fetal immune system become evident in the human at a stage as early as two to three months of gestation. This system must adapt to two hosts, the autologous and the maternal. It is accomplished through the establishment of a delicate balance between mutually contradicting forces. The development of the fetal immune system is as much the result of little understood induction and recognition factors as the development of other major organs and systems.

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Immunoglobulins and Fetal Infection

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Most fetal antibody activity is caused by placental transfer of maternal immunoglobulin G (IgG). However, determination of cord IgM levels is a useful adjunct to the diagnosis of clinically inapparent and atypical intrauterine infections (1). IgM in cord blood is reported to be frequently increased in congenital infections but not in all cases (2). Elevated IgA levels have not been widely applied to the diagnosis of intrauterine infections mainly because of the lack of sensitive methods to measure the IgA in cord blood. We have used a hemagglutination inhibition method in which sheep red cells have been coated with monospecific myeloma proteins using glutaraldehyde (3). We also presented evidence using genetic typing of IgA that the fetus can synthesize IgA in utero (4). Fetal IgA was detected as early as 14 weeks of gestation in cord blood and amniotic fluid. Our data indicate a role for IgD in the developing fetal immune response which may be more closely related to regulation and maturation of the lymphoid system than to humoral antibody elicited by antigenic stimuli (5). IgE does not seem to be involved in the fetal immune response to infection (6).

Our original interests were 1) to determine to what degree IgA and its subclasses IgA₁ and IgA₂ were involved in the immune response of the fetus, 2) to determine if amniotic fluid Ig values reflected the immune response of the fetus and assess their potential value in prenatal diagnosis, 3) to explore the interrelationship between cord blood and amniotic fluid Ig concentrations with respect to the fetal immune response and, 4) to determine fetal factors important in evaluating the data.

Premature Rupture of the Membranes

A preliminary report of 16 patients with prematurely ruptured membranes (PRM) studied in this

laboratory indicated that the fetus showed higher cord blood Ig values in some cases of PRM (6). There was an individual variability in the Ig produced by the fetus when infection occurred: a significant increase in either or both IgA or IgM, or no increase at all was found. We interpreted these changes as a result of immune response to ascending infection from the maternal genital tract. An enlarged series comprising 227 cases of PRM confirmed this report indicating a humoral immune response by the fetus in some but not all patients with PRM (7). It was of a special interest to note that two patterns of infection could be identified when the time interval between PRM and the onset of labor were considered. There was immunologic evidence of a first peak after one to twelve hours of PRM and a second peak could be identified after 72 hours of PRM. The first peak suggested that an infection was present before PRM, and it might have been the direct cause of PRM. The combination of intact membranes and amnionitis is well known (8). A study of Bobitt and Ledger (9) as well as our own study suggest fetal exposure to infectious agents in some patients before the clinical onset of symptomatology of membrane rupture. The second peak was thought to represent the expected increased infection rate after prolonged PRM. We believe, that if the first group could be identified early, the patients should be delivered as soon as possible and should not wait for spontaneous onset of labor at a later time. These studies clearly indicated that IgA determination should also be included in cord blood studies, provided a sensitive method is available. Several infections, however, associated with PRM had normal cord Ig levels. This could be due to either immature immune competence or immune paralysis, but proof is lacking.

Specific Intrauterine Infections

Although cord blood IgM has been used extensively to diagnose intrauterine infections, many

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false negative results have been encountered (9, 11). This is partly because abnormally elevated IgA levels have not been widely used to diagnose intrauterine infections. Mainly, methods sensitive enough for determination of cord blood levels have not been available to most investigators (3). Measurable IgA in normal cord sera was found in 1.5%–33% using single radial diffusion (11–14). IgA was found in measurable amounts in 29.9% of specimens examined using immunoelectrophoresis (15). Using a radioimmunoassay, measurable levels of IgA were found in all specimens tested (16). Our findings are consistent with this last study indicating the feasibility of detecting IgA in cord blood. We used a method in which IgA, its subclasses IgA₁ and IgA₂ were semiquantitatively measured by inhibition of hemagglutination tests of sheep red blood cells coated with monospecific myeloma proteins (3).

Fourteen mothers who contracted toxoplasmosis during pregnancy and their 14 offspring were studied (3). Four newborns developed toxoplasmosis, ten did not. The four infants with congenital toxoplasmosis had evidence of increased immunoglobulin synthesis in utero in sharp contrast to the ten offspring to toxoplasmosis-infected mothers who failed to develop the disease. Three of these four affected infants had elevated IgM levels, while all four infants had significantly increased IgA values. The use of IgA subclasses IgA₁ and IgA₂ was not helpful in distinguishing infants with congenital toxoplasmosis from unaffected infants.

The fetus can respond with accelerated synthesis of immunoglobulins already during the second trimester of pregnancy. One of our patients had amniocentesis twice and was aborted four weeks after the last one at 24-weeks gestation because of Trisomy 21 of the fetus (17). Cultures of fetal splenic tissue were positive for *Propionibacter acnes*. Cord blood IgM was significantly increased for 24 weeks of gestation (19 mg/100 ml).

In a case of congenital rubella, rubella virus was recovered from the amniotic fluid and fetal tissues at 16-weeks gestation after therapeutic abortion was performed (18). The fetus had normal concentrations of IgA and IgM in amniotic fluid. Although IgM was recovered in the amniotic fluid, the anti-rubella titer was nil. This indicates that, although the fetus could produce IgM in normal amounts, it could not make rubella-specific antibodies. The lack of evidence of an immune response by this 16-week-old fetus may be attributed to either immune paralysis or inability of the fetus to produce this specific immune response during 8–16 weeks of gestation. This finding is consistent with the known

ability of the rubella virus to persist in the fetus throughout gestation and early neonatal period.

Transplacental passage of herpes simplex virus has been associated with fetal infection (19). However, we have recently studied a case, in which maternal herpes genitalis was associated with herpes virus in the amniotic fluid without overt fetal infection (20). This case suggests that the presense of herpes simplex virus in the amniotic fluid does not necessarily indicate overt fetal infection. It is possible that the maternal antibodies to herpes simplex virus in this case of recurrent infection were transferred to the fetus through the placenta as IgG (20). The similar titers of antibodies to herpes simplex virus in maternal serum and cord blood suggest that this was the case. It is not unlikely that these antibodies produced by the mother and transferred through the placenta protected the newborn from infection (20).

Fetal Factors Affecting Ig Levels

As immunoglobulin concentrations were studied, it became evident that fetal factors were important in evaluating the data (21). Cord blood IgM and IgG both increase with gestational age until term, while IgA, its subclasses IgA₁ and IgA₂ and IgD were unaffected by gestational age (22). IgM and IgG were higher in fetuses weighing 2,000 grams or more, while IgA, IgA₁ and IgA₂ were not influenced by birth weight. This is important since a cord blood IgM level of 10.5 mg per 100 ml, for example, is significantly elevated in a fetus of 35-weeks gestation or less, and/or a birth weight of 2,000 grams or less, while this cord blood value is considered normal in a fetus of more advanced gestation and/or of a higher birthweight (22). Cord blood values are the same in male and female infants (22).

Newborns with Trisomy 21 have significantly lower IgM and IgA cord blood levels when compared to controls and to congenital disorders of various sorts (17). This suggests that the fetal Ig synthesis is impaired in Trisomy 21 in utero as the consequence of a primary immunodeficiency. In spite of this, fetuses with Trisomy 21 can respond to intrauterine infection with increased synthesis of IgM (17).

Fetal Immunoglobulin Production Following Maternal Immunosuppression

Although immunosuppressive therapy has been given to many pregnant women without demonstrable ill effects on either mother or fetus, there are reports in the literature of premature labor, pre-

mature rupture of the membranes, fetal malformations, adrenocortical failure in the fetus, fetal lymphopenia, and intrauterine growth retardation following maternal immunosuppression (23). We have studied women who received azathioprine and/or prednisone during the entire pregnancy following kidney transplantation or because of systemic lupus erythematosus (23). The fetuses appeared to have intact humoral immune function in that they had normal cord blood Ig concentrations and could respond with increased synthesis of IgA and IgM following premature rupture of the membranes. We have also demonstrated that fetuses of mothers who received dexamethasone or betamethasone at 29 to 36-weeks gestation during three consecutive days to induce maturation of the fetal lung do not differ significantly from normal control subjects (24). In a subsequent study (25), cord IgA and IgM levels were measured in newborns of mothers with prematurely ruptured membranes who also received prenatal glucocorticoid treatment to accelerate fetal lung maturity at 22 to 34-weeks gestation. A mixture of slow and rapid acting glucocorticosteroids showing prolonged activity (four to six weeks) were used. The data showed that the humoral immune response also takes place after the fetus has received antenatal glucocorticoid treatment for a prolonged period of time in that four infants in the betamethasone-treated group ($n = 14$) responded with elevation of IgA and/or IgM after premature rupture of the membranes. Whether or not the fetal immune response is vulnerable under more stringent conditions has to be further evaluated.

Amniotic Fluid Immunoglobulin Levels

Demonstration of elevation of Ig values in amniotic fluid is potentially informative for the diagnosis of intrauterine infections. We have shown that when sensitive detection methods are applied all Ig classes and subclasses can be detected in amniotic fluid (4, 26, 27). In 161 samples tested throughout gestation, IgA, IgA₁, IgA₂ and IgG were measurable in all cases; IgM was measurable in 99.4 percent and IgD in 90.6 percent of the fluids (27). IgA, IgG and IgD concentrations increased toward midpregnancy and thereafter decreased to term in a pattern similar to that for amniotic fluid total protein. IgM remained relatively constant through week 35 of gestation and thereafter it increased to term. The mean IgA values were 2.2 times higher in amniotic fluid than in cord serum. This remarkable finding was in sharp contrast to IgM, which was only 1/16th as high and to IgG, which was only 1/66th as high in amniotic fluid as in cord blood. The data for

IgA as compared to other immunoglobulin classes suggest that amniotic fluid IgA may be partially derived from IgA in fetal gastrointestinal and pulmonary secretions.

Elevated cord blood Ig levels are often reflected in increased amniotic fluid Ig levels (21, 27). An infant born in our hospital to a mother who acquired toxoplasmosis during pregnancy had increased levels of IgM in both cord blood and amniotic fluid, while the IgA levels were normal in both. Another infant born after premature rupture of the membranes had increased levels of IgA but normal levels of IgM in both cord blood and amniotic fluid. An anencephalic fetus, on the other hand, had normal cord blood values for IgA and IgM, while the amniotic fluid had elevated IgM but normal IgA. The elevated IgM in the amniotic fluid of the anencephalic infant is consistent with other reports claiming that IgM leaks out from the meninges into the amniotic fluid (28).

A Look into the Future

Further information on the value of immunoglobulin determination in cord blood and/or amniotic fluid will be available when more data on pathologic pregnancies, such as intrauterine infections, malformations and immunodeficiencies are available. In cases of Ig elevations, IgM can be isolated by column chromatography and IgA by immunoabsorption techniques. The isolated fractions can then be tested for specificity against a panel of infectious agents. This will not only be helpful in establishing a specific diagnosis, but also important in the search for the pathogenesis and prevention of birth defects of unknown origin.

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Immunology of Infertility

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Abstract

Relative infertility may be causally related to the presence of sperm antibodies in a female, male or both. Sperm antibodies may be of agglutinating or immobilizing types, and may be present in the serum, seminal plasma, or cervical mucus.

Since the apparent incidence of such immunologic infertility in infertile couples is about 5%, it seems reasonable to include testing for antibodies in the work up. The laboratory methods most applicable to this problem are the micro- and macroagglutination tests, as well as the sperm immobilization tests.

Once the diagnosis is made, the therapeutic regimens for the affected female are occlusive therapy, and/or immunosuppression. In the case of a male autoimmune response, the treatment may be the sperm-washing-insemination technique or immunosuppression.

The reported pregnancy rates following these methods range from 10% to 60%.

Because of the ease of the laboratory diagnosis of sperm antibodies and their specificity, which is due to the easy elimination of nonspecific beta-agglutinins, we suggest the immunologic work up as a part of a routine investigation of the infertile couple.

Introduction

The true incidence of infertility is difficult to ascertain, but has been estimated at 10% to 15% in the general population (1, 2). Furthermore, approximately 15% of couples will achieve a smaller number of children than desired. In this infertility population, a well-planned and systematic search for etiologic factors consists traditionally of:

- 1) Evaluation of the male partner
 - a) Sperm analysis
- 2) Evaluation of the female partner
 - a) Ovulatory process
 - b) Uterotubal architecture by means of hysterosalpingography
 - c) Evaluation of sperm-cervical mucus interaction by postcoital test (PCT)
 - d) Endoscopic examination of the pelvis

Upon completion of this work up, a precise diagnosis as to the causative factor can be reached in about 80% to 85% of the couples, the remainder being labelled "infertility in a normal couple." For

these couples, as well as for those who exhibited poor postcoital test, in absence of inflammatory or infectious disease process in the genital tract, evaluation of the immunologic factor has usually been recommended. Henceforth, detection of sperm antibodies in either partner added 5% to the group with precise diagnosis. Couples in whom the antibodies can be demonstrated have what we call "immunologic infertility" (3).

It seems reasonable to recommend immunologic evaluation as a part of a routine infertility evaluation, for the following reasons. First, the combination of the causative factors in infertility is common. Second, based upon the findings in our study (4), the results of the postcoital examination do not always correlate with the presence of sperm antibodies. Although the percentage of positive antibody tests increases progressively as the postcoital test results deteriorate, we have found that in a group with an excellent PCT result, 7% of the women and 21% of the men showed the presence of antibodies. The poor results of PCT appear to well reflect the presence of antibodies in the female, but the antibody tests in the male do not correlate well with the clinical findings on PCT examination.

It is important to emphasize that immunologic infertility appears to be relative, and not an absolute factor. The presence of antibodies in either partner

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may be reflected in a lengthy delay in the time required to achieve pregnancy.

The purpose of this paper is to review the available data on the antigen, the antibodies in the infertile couples, to present the methods of detection, and to summarize the available means of therapy.

Antigens

Shulman (1) has classified the antigens of the male reproductive tract into two systems: 1) spermatozoal and testicular antigens and 2) antigens of the accessory glands of reproduction.

Sperm, by definition, are autoantigenic since they are antigenically different from the male producing them. However, since they do not develop until puberty, when the immune system is already fully immunocompetent, they are usually shielded from this system by a blood/testis barrier (5). Theoretically, it appears likely that in certain situations this immunologic barrier may be broken. In experimental animals exposed to autologous sperm, antisperm antibodies are produced, which agglutinate or immobilize sperm.

In humans, sperm antibodies can be detected in the serum of about 2% of fertile and 8% to 13% of infertile men (6-8). Immune orchitis can be experimentally induced in animals (9) and man (10), but it has not been established whether this phenomenon can occur spontaneously in human beings. It has been reported (11-13), that 50% to 60% of men have sperm antibodies within the first year after vasectomy. Thus, it might be postulated that the antigen leakage with resulting autoimmune response may occur as the result of orchitis or testicular injury. Most importantly, the accessory glands, especially the prostate, serve as a major source of immunoglobulins in the ejaculate. At the time of ejaculation, immunoglobulins are added to the seminal plasma, and may affect sperm motility (14).

Li and Behrman (15) reported that there are at least four specific antigens in the seminal plasma. No specific antigen could be identified in the spermatozoa.

Antibodies-Female

Three types of immunologic reactions to sperm antigens may occur:

- 1) The systemic antibody response (16-18)
- 2) The cell-mediated immune response (19, 20)
- 3) The local immune response (detectable mainly in the cervical mucus) (21-23)

Franklin and Dukes (16) initially reported a 72%

incidence of circulating sperm agglutinins in the sera of female patients with unexplained infertility, as compared with a 5.7% incidence in a control group. In their second report (24) they stated that the incidence of sperm agglutinins in the infertile population was only 48%, with a control group showing the incidence of 13%. In the experience of other authors, there has been a great variation in the estimate of the incidence of sperm agglutinins in infertility, ranging from 37.5% (25) to 20% (26). Shulman (27) reported an incidence of positive macroagglutination tests in 19% and microagglutination in 10% of infertile women. His controls were in the 2% to 3% range.

Such widely varying results have caused considerable controversy and doubt. Most importantly, when pregnant women were used as controls, several investigative series showed that the frequency of sperm agglutinins found were comparable or even higher than those found in infertile women (25, 28-30).

Most recent reports (31, 32) have revealed that the agglutination of spermatozoa by the sera of women, as detected by the Franklin-Dukes test, can be caused by both sperm antibodies and by the presence of a high molecular weight compound with beta mobility in preparative zone electrophoresis, called beta-spermagglutinins.

The study (32) indicated that these compounds occur with comparable frequency among sera from infertile and pregnant women, as well as among the users of oral contraceptives. Obviously, this type of agglutinating activity does not appear to be a causative factor in infertility. A close association between female infertility and the presence of circulating sperm agglutinating antibodies was demonstrated.

Sperm immobilizing antibodies appear to belong in a somewhat different category. In the original studies, 19.4% of women with unexplained infertility were found to demonstrate their presence (18, 33), while there was no detectable sperm immobilizing activity in the sera of pregnant women used as controls.

The local immune response in the female-appearance of sperm antibodies in the cervical mucus (21-23)-has been attributed to locally produced antibodies by the cervical mucosa. The possibility of a female cellular immune response against spermatozoal antigens, with resulting reduction of fertility potential, has also been raised (20).

In vitro, cellular migration inhibition test for the detection of cell mediated immunity has been described (34). It has been stated that the leukocyte

migration inhibition test can be used for the detection of such cell mediated immunity against spermatozoal antigens in the infertile women (19).

Thus, cytotoxic antibodies were demonstrated in the cervical mucus, which caused immobilization and cytotoxicity of the sperm (21). It has been demonstrated that immobilizing antibodies reduce sperm motility in the cervical mucus (35, 36).

The female genital tract normally contains immunoglobulins, secreted in the cervical mucus. Although IgM is almost absent, there are abundant amounts of IgG and IgA (37). Interestingly, the ratio of IgG and IgA has been shown to be 3:7, which is not typical for secretions in general (38).

Male Autoimmunity

Autoimmunity as a cause of infertility in men was reported initially in 1954 (39).

Rumke and Helliaga (40) reported significant titers, in excess of 1:32 of sperm agglutinins in 3% of the 2000 husbands of infertile marriages. The controls, fertile men, have shown no agglutination in titers greater than 1:16.

Conclusive evidence of autoimmunity as a cause of male infertility was presented by Fjabrant (41).

Shulman (8) found the incidence of sperm agglutinins in infertile men to be 9% (by macroagglutination) and 5% (by microagglutination).

Antibody activity in a male can be demonstrated by the serum, seminal plasma, or both. Agglutinated spermatozoa are unable to penetrate the cervical mucus. It has been shown that the portion of non-agglutinated spermatozoa in the ejaculate was also often sensitized by the antibody and that these sperm progressively lost mobility as they penetrated the cervical mucus (42).

Both the sperm agglutinating and immobilizing factors in men seem to be mainly antibodies of the 7 S type.

Materials and Methods

The scope of this paper allows for only a brief description of the laboratory methods for detection and diagnosis of sperm antibodies. The laboratory assessment of the infertile couples should include examination of serum from both partners, in addition to the seminal plasma and the cervical mucus. The four methods applicable for the detection of antibodies include agglutination, immobilization, cytotoxicity and immunofluorescence. Only the first two appear to have clinical significance and applicability. The technical details of these methods have been extensively reviewed in other publica-

tions (44, 45). New terminology for these methods has also been proposed (45).

I. Agglutination

A. Gelatin Agglutination Test (GAT), based on Kibrick's technique (46) represents a macroscale procedure (sperm-serum/seminal plasma/cervical mucus), allowing the observation of agglutination in small tubes, and for titration of the sperm antibody activity by study of a dilution series.

B. Tube-Slide Agglutination Test (TSAT), popularized by studies of Franklin and Dukes (16, 24). In this test (sperm-serum/seminal plasma/cervical mucus), a microscopic assessment of free and clumped cells is made. The total number of clumped cells is divided by the total number of all cells, giving a percentage figure. The result is positive only if it gives a percentage higher than 10%.

C. Micro-Agglutination Test (MAT), represents a microscale procedure described by Friberg (47). It is now called Tray Agglutination Test (TAT). It utilizes microsyringes to mix serum and sperm in the areas of a tissue-typing plate. This method has the advantage of the ease of observation of the mode of agglutination-head-to-head, head-to-tail, tip of the tail-to tip of the tail.

D. Slide Agglutination Test (SAT), merely a qualitative method, generally used to supplement the GAT.

II. Immobilization

A. Isojima Sperm Immobilization Test (SIT-I), is based on the fact that some antibodies to spermatozoa can cause the immobilization of sperm cells, as long as the system contains complement (pooled fresh rabbit serum, human or guinea pig serum). By this method, one determines whether the motility in the mixture of serum, sperm and complement has fallen below one-half of the motility in a control mixture. This technique is also called the Complement Dependent Technique.

B. Fjabrant Sperm Immobilization Test (SIT-F), represents a modification of the previously described method.

Additional technical details of all of these methods have been previously reviewed (3, 48).

Therapy of Immunologic Infertility

The effective and ultimate therapeutic method for the couple with immunologic infertility is yet to be found. So far, the longest established procedure in the case of a woman with sperm antibodies has

been the continuous condom therapy, also termed "occlusive treatment." The use of the condom for every coitus prevents the repeated absorption of sperm antigens. The duration of this treatment should be six to 18 months, during which time we recommend assessment of antibodies and their titer every three months. Obviously, every woman is different in her immunologic memory and antibody-decay mechanism; if the antibodies do not disappear or decrease significantly within 12 months of this therapy, they probably will not. If the decrease has occurred, however, we allow the couple unprotected intercourse at the time of ovulation. The success of this method cannot be stated with accuracy, but it has been estimated to be about 40% to 60% accurate (48).

It is of note that the condom therapy is applicable in cases of spermagglutinins only; the titers of sperm immobilizing antibodies do not seem affected by this method (14).

Donor insemination is no value in so affected women, since the sperm antibody is directed against the sperm cells from all men.

Attempts at immunosuppression, using large doses of steroids for a short period of time, has been recommended by Shulman's group (3), and a limited group of women resulted in a success rate of 14%.

When the male demonstrates autoantibodies to sperm, we have utilized the sperm-washing-insemination method (SWIM) (3). It is based on the rationale that the antibody present in the seminal plasma can be washed away. The semen is diluted four-fold with the suspending medium (4% solution of human serum albumin) and centrifuged at 2000 rpm for five minutes. The soft pellet is then resuspended and centrifuged again. This washing procedure is done three times. The final pellet is suspended in about 0.5 ml of the medium and used for the intrauterine insemination. In a limited group of patients, the success rate has been about 10%.

We have also described an immunosuppression method for the male partner (3), consisting of administration of methyl-prednisolone, 96 mgm/day, for seven days. With this method, autoantibody titers may decline as early as 14 days after the onset of therapy, and may remain suppressed for several months (50). Ensuing pregnancy rate has been 35% in our experience (3).

In some instances, when the SWIM method and/or immunosuppressive therapy did not produce the desired result, we have recommended donor inseminations to the couples, with achieved success.

Recently, an additional treatment regimen has been reported (51) consisting of a pharmacologically

induced azoospermia, followed by the administration of corticosteroids. This approach was reported to restore fertility in 18.75% of a group of 48 men.

Conclusions

The presented literature review and our personal experience indicate that, in a limited number of couples with primary or secondary infertility, immunologic factors appear to be the sole factor responsible for their relative reproductive failure. There has been, however, considerable doubt whether sperm agglutinating antibodies really alter the fertility potential, since there have been reports of their presence in the undiluted sera of pregnant women, and patients using oral contraceptives. The description of nonspecific beta-spermagglutinins allows for the explanation of this phenomenon. Apparently, there seems to be an association between high steroid levels and the development of beta-spermagglutinins. The strikingly higher frequency of spermagglutinins in sera from women in advanced pregnancy supports such an hypothesis (32). Thus, these recent observations allow for differentiation between spermagglutinating antibodies, which seem to have an important role in impairment of fertility, and beta-spermagglutinins, which are nonspecific. Ingerslev (32) described the absorption procedure that neutralizes activity due to the nonspecific compounds, allowing a precise diagnosis. For clinical application, it appears that the Kibrick and Franklin-Dukes testing methods (GAT and SAT) give thoroughly reasonable results, when properly performed. Immobilization tests, while very specific to infertility, appear to be less sensitive.

Infertility in men caused by autoagglutination has been well documented. It does not seem to correlate well with the results of PCT (4), unless oligospermia is present. This occurs in about one-third of patients with antisperm antibodies (40, 52). Based on this observation, we recommend immunologic work up for all infertile males with decreased sperm counts. Therapeutic methods for immunologic infertility, both female and male, are still in the process of development.

In the case of the female factor, occlusive therapy and attempts at immunosuppression are the only available methods. In cases of male autoimmunity, intermittent high dose methylprednisolone treatment seems effective in reduction of circulating antibodies. It seems that suppression of IgA in the seminal plasma is the most important factor in freeing sperm of the antibodies which impede penetration of the cervical mucus (53). Our success rate

with this regimen (3), in terms of pregnancies, has been confirmed by others (52).

Although our understanding of the immunologic factors in infertility is still far from complete, their reported incidence, the ease of laboratory diagnosis, and the encouraging results of the therapeutic efforts seems to justify inclusion of diagnostic search for these factors in all infertile couples.

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Studies on Immune Infertility: A Hypothesis on the Etiology of Immune Infertility Based on the Biological Role of Seminal Plasma Immune Response Inhibitor

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Abstract

Normal female does not produce an immune response against "foreign" seminal antigens. During coitus, the female reproductive tract is inoculated with millions of sperm of alien pheno genotype, suspended in a complex proteinaceous medium. Coitus therefore provides a substantial antigen dose for immunization. A hypothesis is presented to explain the failure of immunologic sensitization in spite of repeated "inoculation" by coitus. An inhibitor of immune response was first identified by us and demonstrated by its potent activity in several *in vitro* reactions such as specific polyclonal lymphocyte activation, allogenic stimulation (mixed lymphocyte reaction) and immunization *in vitro* by sheep erythrocytes. This inhibitor is adsorbed along with the other seminal plasma components on the spermatozoa and there confronts the female immunocompetent cells simultaneous with the seminal antigens. The inhibitor interferes with antigen recognition and/or the responsive capacity of immunocompetent cells of the female. The male partner of immune-infertile couples who has either the absence or quantitatively lowered levels of this inhibitor in his semen renders the seminal antigens semen immunogenic for his female partner and thus initiates a series of events resulting in immune infertility.

Introduction

Immune response to antigenic components of spermatozoa and seminal plasma has been well

documented (6, 32) in women and men and a proportion of these patients suffer from infertility.

Normal mammalian females do not produce immune response against antigenic components of semen in spite of fulfillment of ideal conditions required for immunization through coitus. The female during coitus receives in her reproductive tract semen from a genetically dissimilar male. Antigenic components of semen, such as autoantigens, alloantigens, sperm proteins, seminal plasma adsorbed on sperm surface, and seminal plasma which is the carrier of spermatozoa in semen, have been demonstrated to be highly immunogenic when injected into females. Coitus is repeated frequently during adult sex life, thereby providing "reimmunization" when the immunologic system is well developed. On the other hand, the female reproductive tract does not appear to be an immunologically privileged site. Ejaculated spermatozoa do not immunize normal females as a consequence of coitus, while epi-

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didymal spermatozoa have been shown to be immunogenic (5).

Why Do Some Women Develop Immune Response to Semen While Most Do Not?

In discussing the etiology of immune infertility, two areas need to be distinguished to approach the problem scientifically. These are the initiation of immune response (sperm antibody and so far unstudied cell mediated immunity) to seminal antigens, and the effector phase dealing with the mechanisms by which the immune components produce disease.

In this paper, we present a hypothesis to explain the principal mechanism involved in the lack of immune response in normal females and the cause of immune infertility together with the central experimental evidence in support of our hypothesis.

Hypothesis

Our concept of suppression of immunologic response of normal females to seminal antigens and etiology of immune infertility are as follows:

(a) A component of seminal plasma which has been identified as a very potent inhibitor of immune

responses is strongly adsorbed on ejaculated spermatozoa.

(b) When female immunocompetent cells are exposed to seminal antigens, they are also exposed to the action of this inhibitor.

(c) This combination of an inhibitor together with the otherwise immunogenic stimuli interferes with the antigenic recognition and/or with the responsive capability of specific immunocompetent cells of females.

(d) A qualitative or quantitative defect in this inhibitor, controlled by heredity, in the male partner would result in varied degrees of sensitization of the female partner, causing immune infertility in certain cases.

(e) When the immunologic sensitization in female is of the type in terms of target antigen specificity and nature of immune response such as humoral or cell mediated, relevant to the effector phase of the disease and exceeds a certain threshold on quantitative level, the woman suffers from immune infertility.

A diagrammatic representation of the hypothesis is presented in Figure 1. The immunogenicity of epididymal spermatozoa and the lack of immunogenicity of ejaculated spermatozoa is discussed in detail in the following section.

HYPOTHESIS

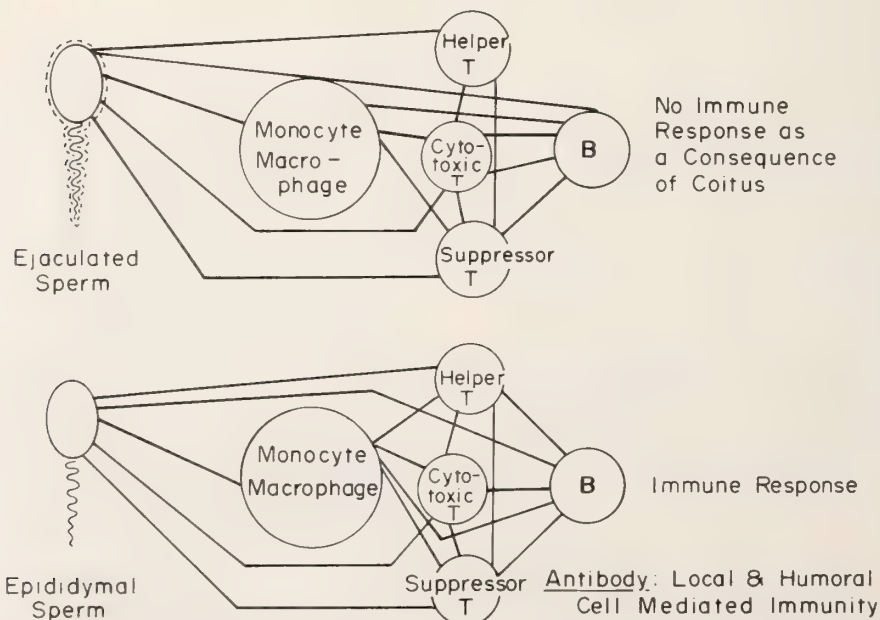


FIG. 1. Our hypothesis presented in a schematic form. Ejaculated sperm by virtue of the absorbed seminal plasma immune response inhibitor (---) is incapable of inducing immune response. Epididymal sperm is devoid of the seminal immune response inhibitor and therefore can elicit immune response in the female reproductive tract.

Possibility of Immunization Via Female Reproductive Tract Through Coitus

Normal females should elicit immune response as a result of repeated coitus but in reality, they do not have detectable immunity to seminal antigens. Coitus from an immunologic view point has four striking features: 1) antigenicity of semen, 2) developmental stage of the lymphoid system of the recipient, 3) frequency and 4) site of insemination for initiating events of immune response.

1) *Seminal Antigens*: Semen has four classes of antigenic components a) autoantigens, b) alloantigens, c) sperm proteins, and d) seminal plasma adsorbed on sperm surfaces as well as seminal plasma itself (6, 52, 74).

Antigenicity of the semen has been established by demonstrating antibodies against sperm and seminal plasma as well as experimental immunization in heterologous and homologous species. Mitchnikoff in 1900 (46) initiated the studies establishing the capability of spermatozoa and seminal plasma components to induce specific autoimmunity and isoimmunity. Meaker in 1922 (43) reported a factor in the serum of two infertile women that was cytotoxic to sperms. There is additional literature on immune infertility (7, 10, 31, 64).

Contemporary literature on the incidence of sperm antibody was reviewed by Beer and Neaves (6). Figure 2 is a diagrammatical representation of the results of nine studies on females and 17 studies on male sera chosen by these authors.

Although spermagglutination is the most widely used test in diagnosis of immune infertility, it is alleged to measure some nonantibody substances in the sera (9, 29). A wide range of false positives from one study to another in the negative controls is strikingly apparent from Figure 2. Apart from the inherent drawbacks in some of the assay procedures, they do not appear to have been applied judiciously in most of the studies on immune infertility. For example, with the proven immunologic technique of immunofluorescence, Maucher et al (42) reported a frequency of positives as 0 out of 40 in their negative controls consisting of fertile non-pregnant and virgin women and a frequency of positives as 6/35 (17%) in unexplained infertility in women. On the other hand, Petrunia et al (51) found as high as 27/48 (56%) positives in their negative controls consisting of pregnant women and 23 out of 51 (45%) positives in unexplained and organic infertility with the same technique of immunofluorescence. In our judgment, this wide dis-

crepancy is due to the lack of required immunologic expertise in the hands of some clinicians. Based on the cumulative data, it is fair to conclude that sperm antibody develop in some women as a consequence of coitus.

Normally, if the immunizing antigen is withdrawn from the recipient, the immune response declines with time. The fact that seminal antigens as a result of coitus provide the immunologic stimuli is substantiated by the observation that after condom therapy in immune infertile patients, sperm antibody titer declines. In the compilation by Beer and Neaves (6), of ten studies ranging from 1967-77 and involving 253 patients, 58.5% showed decrease in the antibody titer or complete absence of antibody in their sera after 6-12 months of occlusive therapy and 31.6% of the patients became pregnant.

Experimental immunization with seminal antigenic components has been successfully attempted in homologous and heterologous species. Ethical considerations in human experimentation prevented further experimentation beyond the first report of Baskin in 1932 (3) when he injected human spermatozoa intramuscularly into a woman and observed subsequent decreases in fertility. Numerous studies on immunization with the semen, washed spermatozoa, seminal plasma, purified components of sperm such as autoantigens, LDH_x, acrosin, etc., and seminal plasma in the same species or different species have established that ejaculated semen undoubtedly has a fairly large number of potent, as well as weak immunogens (23). Alloantigens including histocompatibility antigens have been detected on the spermatozoa of several mammalian species though some of the claims have been disputed (52, 74). Adsorption of seminal plasma on spermatozoa has been demonstrated (49, 75). Antigenicity of seminal plasma components is well documented (16, 22, 36).

2) *Frequency and Site of Deposition of Semen*: During coitus, semen is deposited in the vagina of female in species such as human, macaque, dog, rabbit, cattle, sheep, and in the uterus of species such as rodents, pigs, horses, not far from the cervix. Cervix has been shown to possess the necessary lymphoid system to mount a local immune response against foreign antigens (73). In the normal adult human female, the frequency of mating is two to three times per week for 30 years. Species differences are discussed in several publications (2, 19, 26, 38). Although in some species mating is seasonal, the frequency still meets the ideal requirements for constant reimmunization.

FREQUENCY OF SPERM ANTIBODY DETECTED BY VARIOUS TESTS

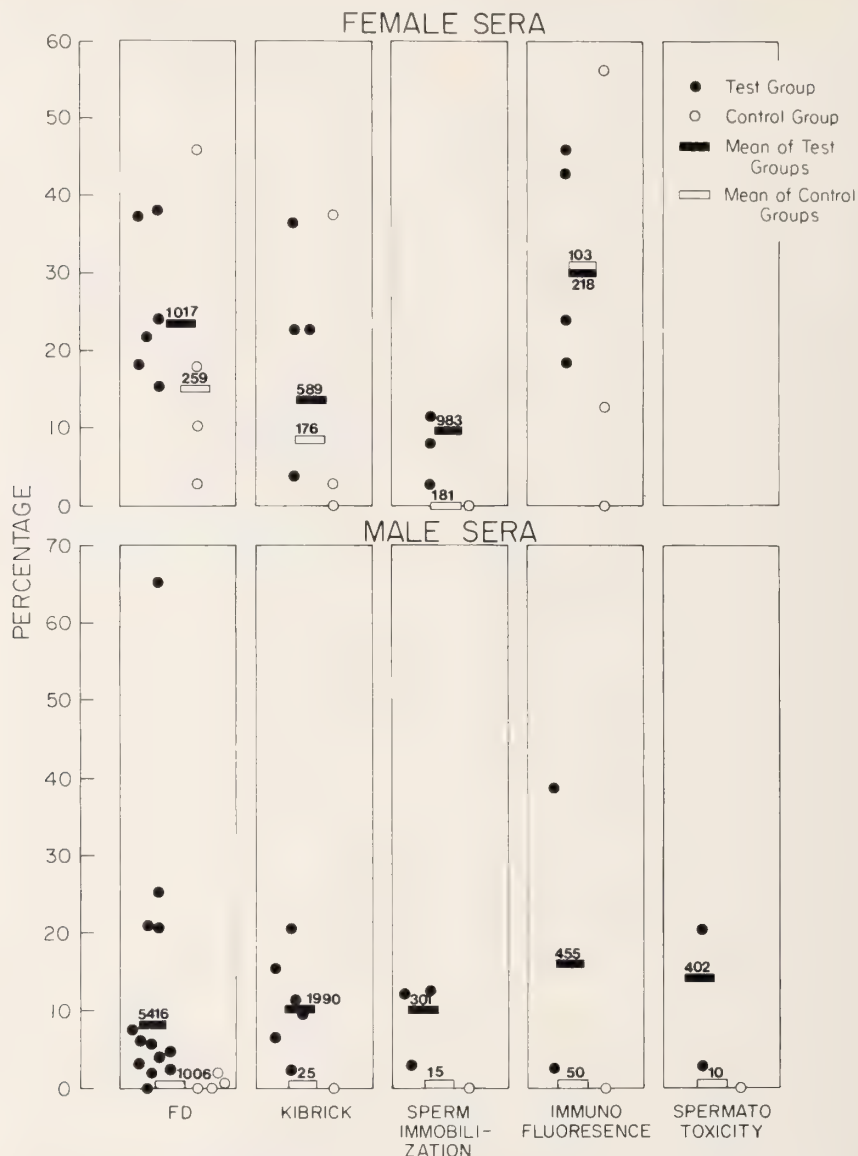


FIG. 2. The figures on the horizontal bar represent the total number in nine studies on females and 17 studies on males for that particular technique. Test group for female (■) is pooled data on positives in unexplained infertility, organic infertility, prostitutes. Control group (□) for female is pooled data on positives in pregnant, fertile nonpregnant and virgins. Test group for male is pooled data on positives in infertile men with azo- or oligospermia, men from infertile marriages, unselected men, men with spermagglutination observed in their semen, men with varicocele, vasectomized men. Control group for male is pooled data on positives in men from fertile marriages. (From Beer and Neaves, 1978)

3) *Recipient's Development of Lymphoid System:* Since coitus takes place in adult life, the lymphoid system is fully developed (68). Therefore the possibilities of neonatal tolerance for suppression of immunity as a consequence of coitus is completely excluded.

4) *Privileged Site:* The "immunologically privileged site" status of the female reproductive tract has been tested by experimental immunization through this route. Capability of cervix to produce

secretory IgA class of antibody locally, and the passage of antigenic information from the reproductive tract to general lymphoid system has been demonstrated.

Austin (2) reviewed the literature on the fate of spermatozoa in the female genital tract. Henderson (27) demonstrated the migration of some leucocyte marked with a carbon label from the uterus to the lymphoid tissue situated antimesometrically within the serous coat of the uterus of rabbits. Reid (59)

studied the fate of ^3H thymidine labelled mouse spermatozoa following coitus. He concluded that some label does reach the regional lymph node via leucocytes or macrophages which phagocytizes the male gametes. The infiltration by macrophages, polymorphonuclear leucocytes and lymphocytes following coitus is well documented (1, 2) and we have ourselves studied this using electron microscopic methods in rabbits. Figure 3 shows a representative polymorphonuclear leucocyte which has engulfed a spermatozoon head. The acrosome, a very thin surface coat and adsorbed material (seminal plasma) on the surface of the spermatozoa are clearly visible. Figure 4 shows several heads of spermatozoa in the macrophage. The nucleus of spermatozoa is distinguishable within the cell. Chemotactic attraction of semen for macrophages and neutrophils was demonstrated by Wilkinson et al (76). We interpret these cumulative findings as providing evidence of the possibility for the passage of antigenic information from female reproductive tract to the lymphoid system of the body.

Evidence for local immunologic response to natural infection or artificially applied antigens in the genital tract of human and other mammalian spe-

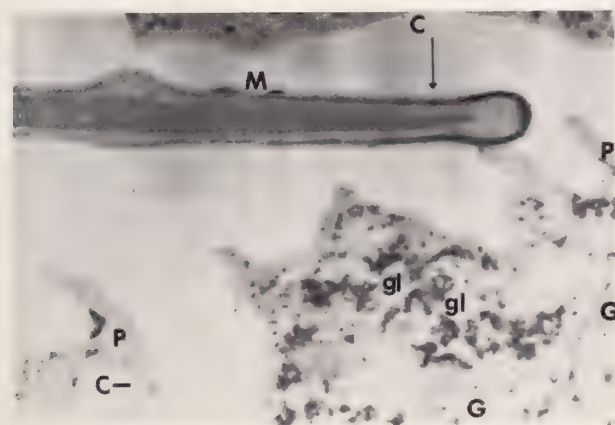


FIG. 3. Uterine contents of rabbits were flushed at 12 hours after coitus. Perfused contents were subjected to fixation by a solution of 3.3% glutaraldehyde in 0.8 M phosphate buffer. After gentle centrifugation, fixation was pursued in a fresh solution of the same fixative for one hour. The cells were finally sedimented by centrifugation. Sedimented cells were embedded in glycol-methacrylate according to the method of Leduc and Bernhard (34). The sections were stained with 1% phosphotungstic acid (PTA) in 1 N Hydrochloric Acid (HCl) at pH 0.3 to specifically contrast polysaccharides and glycoproteins according to the method of Rambourg (58). This picture shows a Polymorphonuclear leucocyte (PMN) cell. The cytoplasm of PMN contains granules (G) and glycogen (gl) which are strongly stained by PTA/HCl. Discontinuous glycoproteic coat (C) is visible on whole surface comprising pseudopods (P). Spermatozoon head is engulfed by PMN leucocyte. PTA/HCl has stained a superficial layer in the anterior segment of the acrosome, a very thin surface coat ($\rightarrow$) and adsorbed material (M) which is likely to be seminal plasma. $\times 30,000$.

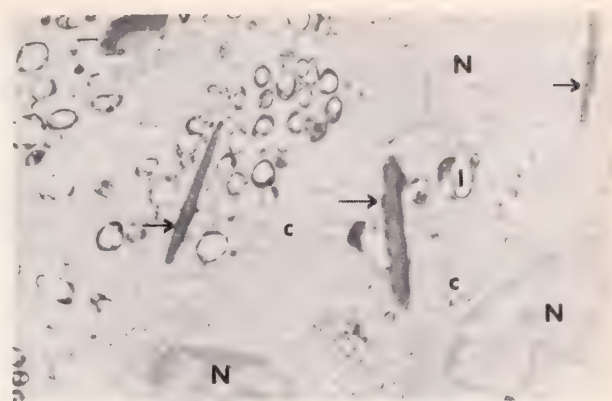


FIG. 4. Uterine contents of rabbits after coitus were processed as described in Fig. 3. Several heads of spermatozoa ($\rightarrow$) in macrophages. Only the nucleus of the gametes is recognizable surrounded by material stained with PTA/HCl. Macrophages shown in the picture have a relatively regular nucleus (N) and stained inclusions (I) and surface (c). $\times 8,000$.

cies has been compiled in a World Health Organization Workshop (18) and the summary is reproduced in Table I. Careful perusal of Table I indicates strong evidence in favor of local immune response. The possibility of rapid degradation of immunogenicity by the proteolytic and other enzymes of the reproductive tract has been excluded from the results of experimental immunization through intravaginal and intrauterine route (5, 8, 24, 77). Schumacher (63) has recently reviewed humoral immune factors in the female reproductive tract.

Beer and Billingham (5) studied the immunization of rodents via intrauterine route. In this species, coitus is intrauterine. Table II shows the effect of immunization of female virgin mice with epididymal sperm in a dose dependent manner through intrauterine route. The assay procedure for immune response was median survival time of test grafts. From their results in Table II, it is apparent that epididymal spermatozoa are immunogenic because of their capability to reduce the survival time of test skin graft. By the same criteria, ejaculated spermatozoa are not immunogenic.

The same authors also studied intrauterine immunization in rats with lymph node cells and epididymal spermatozoa in a dose dependent manner. The intensity of resultant immune response was measured by the weight of paraaortic lymph nodes which drain the uterus and the survival time of test skin grafts. Table III shows the results of this study. The response to epididymal sperm immunization was found comparable to the same number of lymph node cells by the weight of the draining lymph node assay and survival time of test skin

graft. We were encouraged to develop the hypothesis presented herein from the studies of Beer and Billingham (4), and particularly the difference in the immunogenicity of the epididymal and ejaculated spermatozoa of the rodents.

Sabin (61) advised us to consider the implication of the needle path left in several studies accounting for the passage for the antigenic information from the site of deposition of the antigen in the female reproductive tract to the lymph node. We designed a system to test the validity of "privileged site

concept." In our system, the recipients were female virgin mice of several inbred strains. The site of deposition of immunogen was the anterior part of the vagina. Very small volumes (10–20 μ l) containing the required optimal dose of immunizing agents were introduced employing Eppendorf pipette system, thus avoiding all possibilities of rupturing or scarring any epithelial lining or spilling. This was planned with the aim of simulating coitus from an immunologic viewpoint. The material for immunization was chosen with the requirements that the

TABLE I

Evidence for Local Immunologic Response to Natural Infection or Artificially Applied Antigens in the Genital Tract of Human and Other Mammalian Species

Antigen and Type	Species	Site Tested	Evidence of Local Ab Response		
			In vivo Local Ab Response IgA	High Ratios Genital/Serum Antibody	In vitro IgA Antibody Synthesis
Bacterial					
Agglutinin to <i>E. coli</i>	Human	Cervix	—	+	—
Salmonella agglutinins	Human	Cervix-vagina	—	+	—
Brucella agglutinins	Cattle	Uterus-vagina	—	+	—
Diphtheria neut. Ab.	Rabbit	Vagina	—	+	—
Diphtheria hemagglutinin	Rabbit	Vagina-cervix	—		+
<i>Vibrio fetus</i>	Cattle	Vagina		+	—
<i>N. gonococcus</i>	Human	Semen	+	—	—
Viral					
Poliovirus	Human	Cervix-vagina	+	+	—
Herpes simplex	Human	Cervix-vagina	+	+	—
Cytomegalovirus	Human	Cervix-vagina	+	—	—
Other Microorganisms					
<i>Trichomonas</i> sp.	Cattle	Vagina-uterus	—	+	—
<i>Candida albicans</i>	Human	Cervix-vagina	+	+	—
Seminal Fluid					
Sperm cytotoxicity	Human	Cervix	—	+	—
Seminal plasma	Human	Cervix	+	+	—
Sperm immobilizing Ab.	Human	Cervix		+	
Anti-sperm, inhibitory to fertilization	Rabbit	Uterus	+	+	—
Other Proteins					
Inhibitor of blastula development	Rabbit	Uterus	+	+	—
Keyhole-Limpot hemocyanin	Cattle	Cervix	—	+	—
Ferritin	Human	Endometrium	—	—	—
Bovine serum albumin	Human	Cervix	—	—	—

+ Present; — absent or not indicated; IgA includes both 7S and secretory (11S) IgA, From Cinder and deWerk, (1976).

TABLE II

Comparison of the Immunogenicity of Epididymal Versus Ejaculated Spermatozoa

Cellular Inoculum & Dosage	Host Status	Number	Median Survival Time + SD (days) of Test Skin Grafts
Control	Virgin	20	25.0 $\pm$ 1.60
Ejaculated sperm (10–50 $\times$ 10 ⁶)	Virgin*	10	27.0 $\pm$ 1.20
Epididymal sperm-1 $\times$ 10 ⁶	Virgin	12	18.0 $\pm$ 1.50
Epididymal sperm-5 $\times$ 10 ⁶	Virgin	12	16.5 $\pm$ 1.48
Epididymal sperm-10 $\times$ 10 ⁶	Virgin	15	14.0 $\pm$ 1.45

* The cervix was ligated 1–2 h after mating. The recipient were adult C57BL/6 female mice. Sperm were from C57 males. The test skin grafts were from C57BL/6 males. From Beer and Billingham (1974).

TABLE III
Influence of Intra-uterine Inoculation of Rats on Immune Response

Cellular Inoculum		Lymph Node Assay Mean Weight of Para-Aortic		Skin Allograft Assay
Type	Dosage in Million	Syngeneic Cells (Controls)	Allogeneic Cells	Survival Times of Test Skin Grafts MSTs $\pm$ SD* (days)
None (controls)	—	—	—	11.6 $\pm$ 0.3
Lymph node cells	50	7.8 $\pm$ 2.9	28.5 $\pm$ 2.9	6.25 $\pm$ 0.46
	10	9.1 $\pm$ 4.1	17.5 $\pm$ 6.2	6.62 $\pm$ 0.91
	1	—	—	8.5 $\pm$ 1.05
	0.5	5.4 $\pm$ 3.0	18.6 $\pm$ 3.4	11.63 $\pm$ 1.69
Sperm	50	9.3 $\pm$ 4.3	33.2 $\pm$ 18.5	6.6 $\pm$ 0.69
	10	10.8 $\pm$ 3.3	23.0 $\pm$ 6.0	7.75 $\pm$ 0.71
Epididymal	1	6.3 $\pm$ 2.5	20.3 $\pm$ 5.7	9.43 $\pm$ 0.79
	1	Inoculated into foot pads (control)		11.3 $\pm$ 1.18
	10	Inoculated into foot pads (control)		12.0 $\pm$ 2.3

* Medium survival time $\pm$ standard deviation. From Beer and Billingham (1974).

immune response could be estimated in quantitative terms precisely in the assay procedures. Spermatozoa were rejected for inclusion in this study as immunizing agent. Model antigens and other agents along with their assay systems used so far are 1) sheep erythrocytes and Jerne's plaque technique, 2) sheep erythrocytes mixed with lipopolysaccharide (for synergistic effects) and Jerne's plaque technique, 3) T-cell mitogen concanavalin A and B-cell mitogen lipopolysaccharide and uptake of DNA precursors by regional (paraaortic) lymph node cells. For studying secondary response, sheep erythrocytes were injected intraperitoneally followed by deposition in the vagina after a lapse of time. The kinetics of immune response was followed. The preliminary results from these experiments clearly indicate that the immunization can be achieved through the route of the female reproductive tract (56).

Yang and Schumacher in 1979 (77) published the results of their long-term (two year) immunization studies on rhesus monkey. A method of vaginal immunization presented in Figure 5 was developed to simulate coitus. Their method avoided any kind of damage to epithelial lining or the complications of needle path implicated in some of the earlier studies. Since immune response to spermatozoa is difficult to quantitate precisely with the known methodology, they chose two model antigens—Lipopolysaccharide (LPS) and T-4 Coliphages. Vaginal immunization was compared to systemic immunization in their study. Figure 6 shows the results after vaginal application in a monkey and Figure 7 shows the results for comparison after systemic immunization in another monkey. The antibody level in cervical secretions in three out of four

monkeys was higher than that in circulating blood after vaginal immunization. Systemic immunization in comparison resulted in high levels of circulating antibodies, but less than 10% appeared in cervical secretions. At the midcycle, the antibody levels were always low in cervical secretions with both routes of immunization. Oviducts' fluids were recently studied in some of these monkeys and demonstrated the same pattern at the time of ovulation (62). McDermont et al (41) have recently confirmed that the hormonal changes during estrous cycle alter the local mucosal immune response in the genital tract of mouse using an adoptive transfer system of ^3H thymidine labeled lymph node cells.

It appears reasonable to conclude from the cumulative earlier evidence and particularly recent Yang and Schumacher's (77) studies that female reproductive tract is not an immunologically privileged site. The female usually mounts an immune response against any antigen introduced through this route though the magnitude of immune response is comparatively weaker than the systemic route.

Direct Experimental Evidence in Support of the Hypothesis

If female reproductive tract is not a privileged site and sperm and seminal plasma components are immunogenic, the possibility of finding a potent agent for suppression of immune response within the ejaculate was a very logical possibility which we inadvertently discovered in 1970 (53). We were attempting to stimulate allogenic lymphocytes with spermatozoa in vitro and had added in the positive controls (allogenic lymphocyte stimulation) dou-

VAGINAL APPLICATION OF ANTIGENS IN THE RHESUS MONKEY

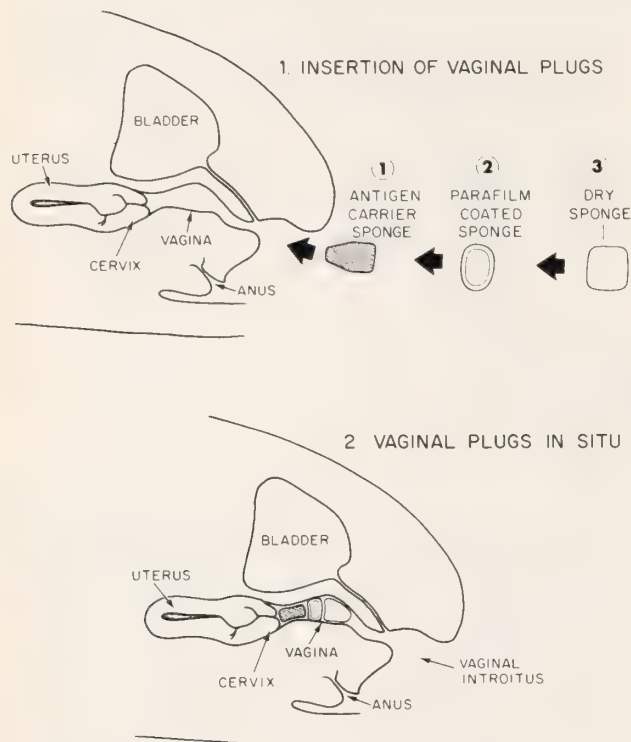


FIG. 5. Dispo plug plastic sponge (1.5 × 2.0 cm cylinder) was filled by injection of 1 ml of antigen solution containing 10^{12} plaque-forming units of phage particles or 5 mg of lipopolysaccharide with or without antigen for each application and placed in the upper part of the vagina against the ectocervix. Subsequently, a sponge covered with Parafilm and a plain sponge were introduced into the vagina to hold the antigen carrier in its position and to prevent leakage. (From Yang and Schumacher, 1979)

bling dilutions of seminal plasma. These positive controls which are representative of T lymphocyte responses were very strongly inhibited by seminal plasma.

This finding suggested that a seminal plasma immune response inhibitor is tightly adsorbed to spermatozoa along with other seminal plasma components and, in this way, confronts the female immunocompetent lymphoid cells that would recognize the male seminal antigens as nonself. This presentation of an "inhibitor" together with the otherwise immunogenic stimuli would possibly interfere with antigen recognition and/or with the antigen responsive capability of the specific immunocompetent lymphoid cells resulting in lack of immune response against seminal antigens deposited during coitus repeatedly in sex life (53, 54). The speculation in this conceptional framework is that males in true immune infertile marriages have either the absence or lower than threshold amounts

of "immune response inhibitor" in their semen, thereby leaving the antigenic components of their ejaculate immunogenic to their female partners.

The experimental evidence favoring the presence of "inhibitor" in the seminal plasma and some of its properties comes from the primary author's earlier published and unpublished studies (53-57) which are summarized in the following section.

In order to establish the presence of a putative inhibitor of immune response unequivocally, the rationale of our methodology was to inhibit immune responses in well defined systems. After excluding the species specificity of the inhibitor, we concentrated on seminal plasma from bulls of proven fertility which were easily accessible. Lymphoid cells were taken from mice since this species has been explored thoroughly by immunologists and certain baseline parameters are well established.

1. The existence of immune response inhibitors in the seminal plasma of human, bovine, swine, and buffalo species was demonstrated in the homologous and heterologous systems of *in vitro* lymphocyte blast transformation by mitogens such as Phytohemagglutinin (PHA) and by allogenic stimulation. Table IV is a representative experiment which clearly establishes the inhibitory effect of bovine seminal plasma on bovine mixed lymphocyte culture (MLC) reaction. The inhibitory activity was abrogated by autoclaving the seminal plasma. Several bull's seminal plasma was found toxic to bovine lymphocytes in certain combinations. We did not pursue the nature of this lymphotoxins in the seminal plasma but made sure in choosing the donors that nonspecific lymphocytotoxicity was excluded as a possible explanation for inhibition of allogenic or mitogen induced responses. From a comparison of 17 bulls, it was evident that individual seminal plasma varies in its inhibitory effect.

2. Though the inhibitors do not show species specificity, other components of seminal plasma can sometimes also produce a strong nonspecific cytotoxicity to lymphoid cells within and across the species barrier, therefore, removal of these by at least partial purification and inclusion of lymphocytotoxicity controls are desirable. Table V shows the lack of lymphocytotoxicity of a discrete fraction of seminal plasma to lymphocytes in heterologous system. The results of several experiments in which the viability of normal mouse spleen cells was assayed by culture in the presence of the highest concentration of Fraction A used in subsequent experiments are shown in Table V. Fraction A is the exclusion peak (tested by blue dextran) of Sephadex G-100 chromatography of bovine seminal

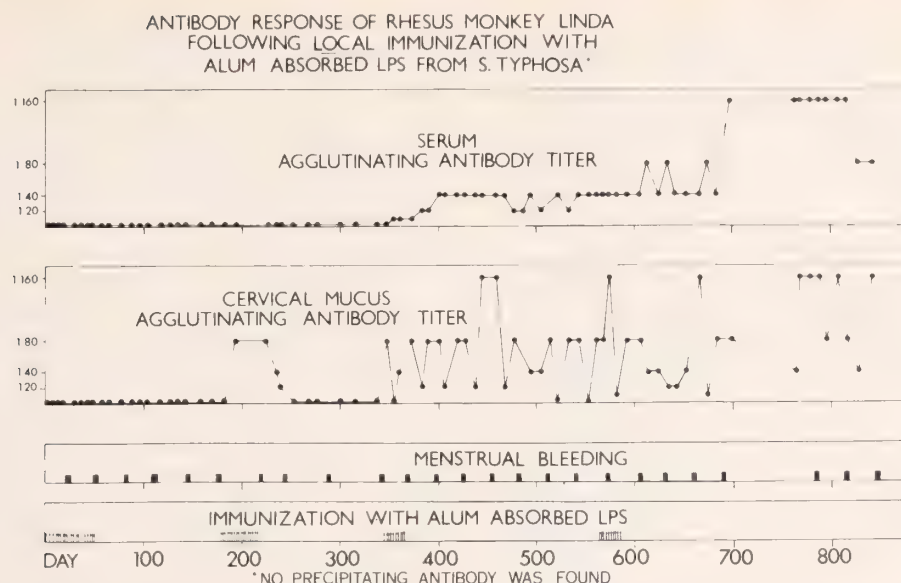


FIG. 6. Each arrow (bottom) indicates one application of antigen. Antibodies to lipopolysaccharide were determined by a modified bacterial tube agglutination test using a commercial preparation of typhoid O somatic antigen. The titer was determined in cervical mucus extracts in consideration of the dilution factor of the extraction. See also Fig. 5. (From Yang and Schumacher, 1979)

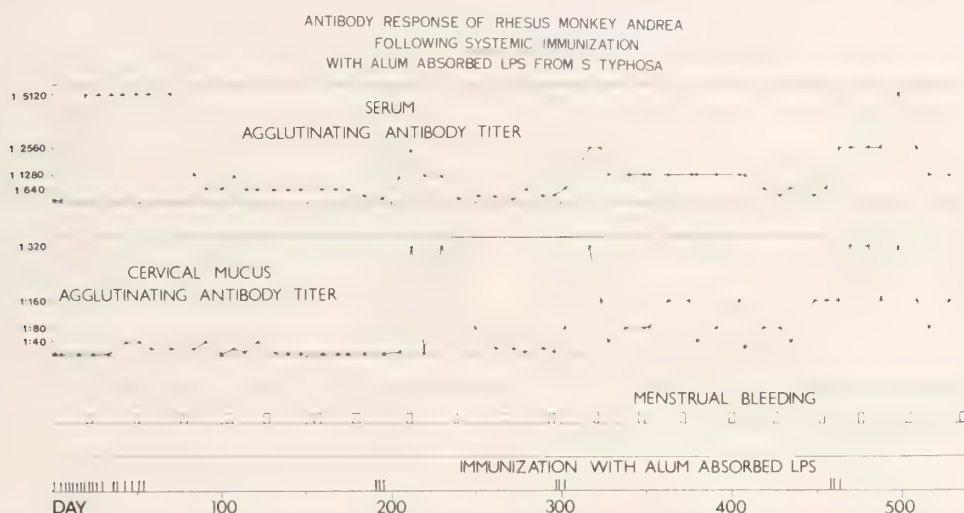


FIG. 7. Each arrow indicates one antigen injection by subcutaneous route. See also Fig. 6. (From Yang and Schumacher, 1979)

plasma. The next peak was highly cytotoxic to mouse lymphocytes. To conclude that Fraction A inhibits cell activation or proliferation, or both, it was necessary to exclude the possibility of cytotoxic effects of these protein concentrations on lymphocytes in culture. Evidently, even these high concentrations of the material did not reduce cell survival in cultures. We therefore conclude that the inhibitory effects were not due to plain cytotoxicity of the fraction. The cell survival was identical with fraction A to that of controls both in serum free and in serum fetal calf supplemented media even during prolonged culture periods (five days) and that the Con A induced responses were measured after only

two days in culture and sheep erythrocytes induced responses were measured after four days.

3. This inhibitor is effective in suppressing the specific immune response or activation in the following systems which we have employed so far:

- a) Specific primary immune response of mice spleen cells against a thymus dependent antigen in an in vitro system quantitated precisely by Jerne plaque technique as shown in Figure 8. Chan et al (14) demonstrated that sheep erythrocytes are thymus (T) cell-dependent antigen. Figure 8 shows that very low concentrations of protein ($7.5 \mu\text{g/ml}$) were able to significantly inhibit the antibody re-

TABLE IV

Inhibition of Bovine Mixed Lymphocyte Culture by Bovine Seminal Plasma

Lymphocytes	Seminal Plasma Final Dilution	CPM Mean $\pm$ SD
Am + A	—	125 $\pm$ 28
Bm + A	—	2,148 $\pm$ 594
Bm + A	1/1100	280 $\pm$ 147
Bm + A	1/1100 heated to 125°-15 min	2,119 $\pm$ 511

Triplicate cultures were performed. The seminal plasma was from a healthy bull of Normand breed two-years-old with proven fertility by artificial insemination data. Seminal plasma was separated from spermatozoa within 4 hours after collection by centrifugation at 600 g for 10 minutes. The supernatant was recentrifuged at 3,000 g for 10 minutes and finally sterilized by Millipore filtration (0.45 μ). The seminal plasma was stored frozen at minus 20°C until use. Repeated freezing and thawing was not found to have any noticeable difference on the activity. Mixed lymphocyte reaction (MLR) was generated in the tube with a total volume of 1.0 ml of Mac-coy medium supplemented with 10% autologous serum from responding donor and 50 I.U. of penicillin and 50 I.U. of streptomycin per ml and buffered with sodium bicarbonate. One million mononuclear cells from ficoll-hypaque density gradient from each of the stimulating and responding donors were mixed. Stimulating donor's cells were treated with mitomycin C (m) at 37°C for 20 minutes (25 $\times$ 10⁶/ml medium 25 μ g of mitomycin C) followed by three washings. Seminal plasma was added in 0.1 ml volume at the beginning of the culture. Several combinations of donors of seminal plasma to lymphocytes were found toxic. Therefore, donors were chosen after screening with eosin viability test. Heating of seminal plasma was by autoclaving which completely destroys the inhibitory activity. The cultures were incubated at 37°C for 120 days in 5% CO₂ in air. MLR was measured by a pulse of 2.0 μ Ci of ³H tritiated thymidine (15 Ci/mole) for 5 hours and harvested on Whatman filter paper.

sponses. Inhibition increased with increasing concentrations of the protein. The cell survival in these cultures was also assessed and was found identical to that of noninhibited control cultures. Therefore, inhibition of the T-cell dependent in vitro antibody response to sheep erythrocytes suggests T-cell suppression as the mechanism of action.

- b) Inhibition of B lymphocyte responses against the hapten NNP (4-hydroxyl-3-5-dinitrophenyl) conjugated sheep erythrocytes induced by polyclonal B lymphocyte activators in fetal calf serum (FCS) is shown also in Figure 8. Bullock and Moller (12) and Coutinho et al (20) showed that substances present in FCS which are mitogenic to B cells activate mouse spleen cells in vitro to polyclonal antibody secretion. This polyclonal activation can be measured by enumerating plaque forming cells (PFC) against erythrocytes coated with a high density of haptenic

determinants. Figure 8 shows the results of inhibition in this system. It can be seen that fraction A was able to significantly inhibit these B cell responses, directly induced by FCS in the absence of antigen. Therefore, it appears more likely that the B cells are also targets for the inhibitory activity of seminal plasma.

- c) Inhibition of T lymphocyte activation by concanavalin A (Con A) in mouse is shown in Figure 9. We used the entire range of effective Con A concentrations in order to distinguish inhibitory effects resulting from inactivation of the mitogen from inhibitory effects on cell activation or proliferation or both. Two dif-

TABLE V
*Lack of Cytotoxicity of Fraction A**

Expt.	Mouse Strain	Fraction A (μ g/ml)	Percentage of recovered viable cells at		
			24 hr.	48 hr.	120 hr.
1	CBA	100	53.8	34.6	14.5
		—	59.6	35.9	15.2
2	(A $\times$ CBA) F ₁	100	50.7	41.5	14.3
		—	49.9	42.2	18.2

* Triplicates of 2 $\times$ 10⁶ viable spleen cells from various strains were cultured in round-bottomed tubes in serum-free medium in the presence of indicated concentrations of fraction A. The number of viable cells in these cultures was counted at the indicated time. Results are given as the percentage of the total number of viable cells put in cultures at zero time. Fraction A is the exclusion peak of Sephadex G-100 chromatography (above 100,000 daltons) of bovine seminal plasma.

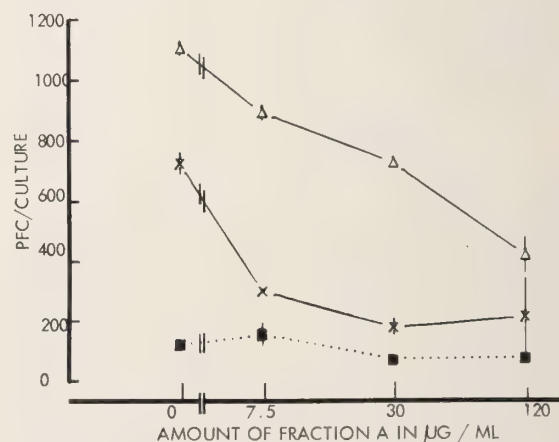


FIG. 8. C57BL mouse spleen cells were cultured (10⁷ cells/culture in 1-ml aliquots) in fetal calf serum-supplemented medium for four days in the presence of indicated concentrations of Fraction A. Half the cultures were primed with sheep erythrocytes (SRBC) and tested for plaque-forming cells (PFC) against SRBC (x — x). The remaining cultures were not primed with SRBC and were tested for PFC against SRBC (■ --- ■) and against NNP-coupled SRBC (▲ — ▲).

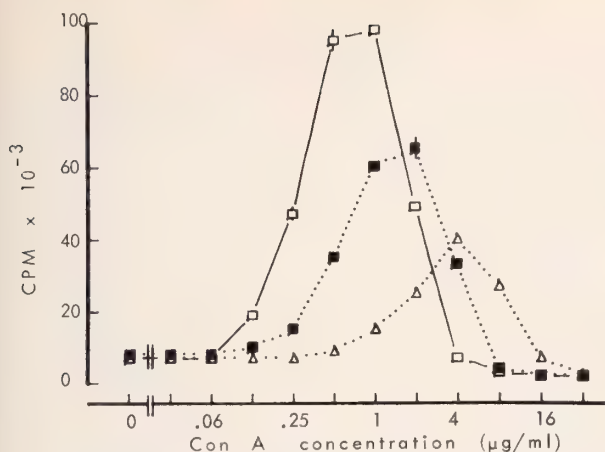


FIG. 9. Inhibition of concanavalin A-induced responses by seminal plasma fraction A. CBA normal spleen cells were stimulated by the indicated Con A concentrations. Cells were cultured either with Con A alone (□—□) or in the presence of 5 µg (■—■) or 20 µg (△—△) of fraction A. 0.5×10^6 spleen cells were stimulated in serum free medium. Fraction A is the exclusion peak from Sephadex G-100 chromatography of seminal plasma (above 100,000 daltons) and was added at the beginning of culture. Cultures were harvested on Day 2 after a 24 hour pulse of ^3H thymidine (Specific activity 5 Ci/mM). Results are shown as mean $\pm$ S.E. of triplicate cultures. The medium was Eagle's minimal essential medium in Earle's solution (Flow Labs, Irvine, Scotland) containing glutamine (2 mM), 100 IU of penicillin and 100 µg of streptomycin/ml at pH 7.0–7.2. Quadruplicate cultures were incubated in plastic boxes filled with a mixture of 10% CO_2 , 83% N_2 , and 7% O_2 .

ferent concentrations of Fraction A from seminal plasma at 5 and 20 µg/culture were tested in quadruplicate cultures. These low protein concentrations of Fraction A effectively inhibited the lymphocyte responses as compared with inhibitory concentrations of whole seminal plasma. The dose-response profile was shifted to the right in the presence of the inhibitory proteins, which suggests that some Con A is inactivated, possibly by binding of the active sites of Con A to carbohydrate residues in the proteins. However, it is also evident that the response never reached the maximal values obtained in the absence of the inhibitor, as would be expected if a mere inactivation of the mitogen were taking place. Furthermore, the degree of inhibition increased with higher concentrations of the seminal plasma fraction A reaching levels of about 30% of control responses.

In view of mushrooming of reports in recent years on putative inhibitors of immune response in biological fluids based on inhibition of lymphoblast transformation (LBT) at optimal concentration only, we would therefore discuss the distinction between spurious and

true inhibition. Figure 10 illustrates diagrammatically dose response curve of the mitogen at suboptimal, optimal and superoptimal concentrations. The response is a normal curve distribution. It is compared in Figure 10 with a) substance under test that binds to the mitogen, b) a lymphotoxic substance, and c) a true inhibitor of mitogen response. Con A responses are used as an example.

If the substance under test binds the mitogen, the maximum response is shifted to the right but the peak response is identical to that without the substance under test. In fact, a number of substances bind to mitogens particularly glycoproteins to Con A. Coutinho et al (20) have demonstrated experimentally

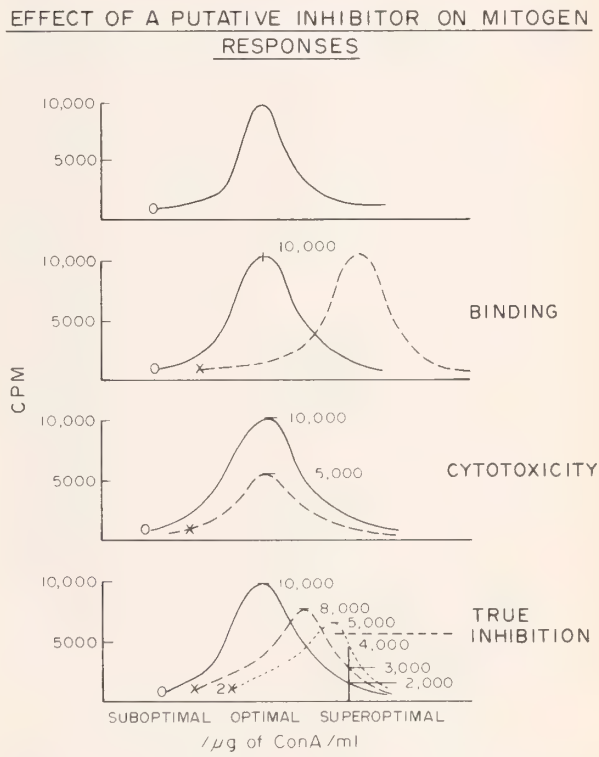


FIG. 10. Hypothetical Con A responses are shown in (○—). The effect of a putative inhibitor on Con A responses are shown in (x---) and at double the concentration of the inhibitor in (2x---). If the putative inhibitor merely binds to the mitogen, the requirements of mitogen for optimal response is only changed to a higher level. However, if the substance under test for inhibitory activity is cytotoxic to lymphocytes, the CPM decreases. In the situation of true inhibition of mitogen response, the response may be shifted slightly to the right if some binding between inhibitor and mitogen occurs but the maximum response in the presence of the inhibitor is always lower than without the inhibitor and are further dose dependent on the amounts of inhibitor. The inhibition is much more at suboptimal as compared to superoptimal amounts of mitogen. Therefore at super-optimal amounts of mitogen the inhibition is either minimal or actually there is enhancement in the case of true inhibitor as shown in the bottom part of the figure.

that the optimal requirement of Con A in serum free medium for maximal response is $1.0 \mu\text{g/ml}$ and in medium containing serum this is augmented to $10.0 \mu\text{g/ml}$. If only optimal concentration of mitogen is used when maximal response is obtained and a putative inhibitor is added, the response can be depressed. This could lead to the conclusion that the substance under test is an inhibitor, but in reality, it can be explained on mere binding. This fact can be borne out easily if the whole range of effective Con A concentration is employed in the experiment. If the substance under test is cytotoxic to the lymphocytes in LBT assay the dose response curve has the same pattern as control but the maximal response is lower and the patterns are similar. In case of true inhibition, even if some binding occurs, the dose response curve may be shifted to right but the maximal response with the substance under test is always lower at any concentration of the mitogen than control and is directly proportional to increasing amounts of inhibitor. In fact, the CPM values with the inhibitor under test are lower than control at suboptimal and optimal concentrations of the mitogen. The curve may be shifted slightly to the right owing to binding of some mitogen however the maximum response with the inhibitor at any concentration of mitogen is lower than the maximum response obtained at the optimal concentration of the mitogen alone. Further increasing amounts of inhibitor should give more depressed responses. Therefore, at suboptimal and optimal concentrations of mitogen, responses with inhibitors are lower than without and at superoptimal concentration of mitogen, it is inverse. Also, the differences in the ascending part of the peak are greater than the descending part of the peak in the absence of binding.

The above effects are the cumulative effects on individual lymphocytes. Figure 11 shows diagrammatically the effect on a single lymphocyte of mitogen alone and with a true inhibitor at suboptimal, optimal and superoptimal concentrations of the mitogen. High affinity cells are stimulated at suboptimal doses of Con A and are paralysed at superoptimal concentrations of the mitogen. Another concept presented in Figure 11 is that optimal number of receptors (not all) need to be triggered to obtain maximal response. At superoptimal concentrations of mitogen with true inhibitor,

MECHANISM OF ACTION OF SEMINAL PLASMA ON CON A ACTIVATION

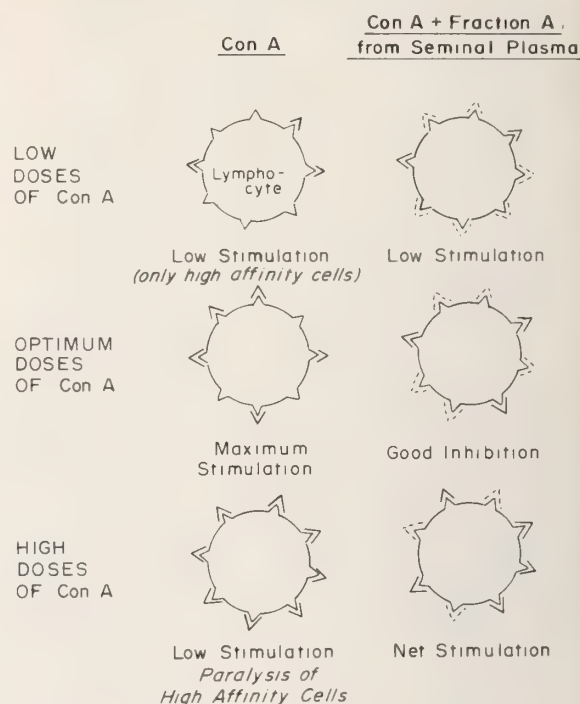


FIG. 11. The effect of varying amounts of mitogen Con A ($\wedge$) on a single lymphocyte is shown along with the effect of a true inhibitor ($\wedge$). The cumulative effects of several lymphocytes generally employed in the mitogen stimulated cultures is shown in figure 10.

the number of receptors on lymphocytes which are triggered are lower than the total number of mitogen receptors and thereby give net stimulation. This could explain why at superoptimal levels of mitogen the response is either parallel or higher with inhibitor than without. Some mitogens such as Con A give a narrow dose response curve while others such as lipopolysaccharide give more broad response.

These strict criterion of true inhibition in mitogen response are fulfilled by fraction A of seminal plasma for both Con A induced T lymphocyte responses and LPS induced B lymphocyte responses.

Actually, another fraction which elutes immediately after Fraction A on Sephadex G-100 chromatography of seminal plasma provides example of cytotoxicity to lymphocytes and we have obtained a dose response curve similar to the one predicted in Figure 10.

d) Inhibition of B lymphocyte activation in mouse by lipopolysaccharides by Fraction A is shown in Figure 12. Fraction A was not

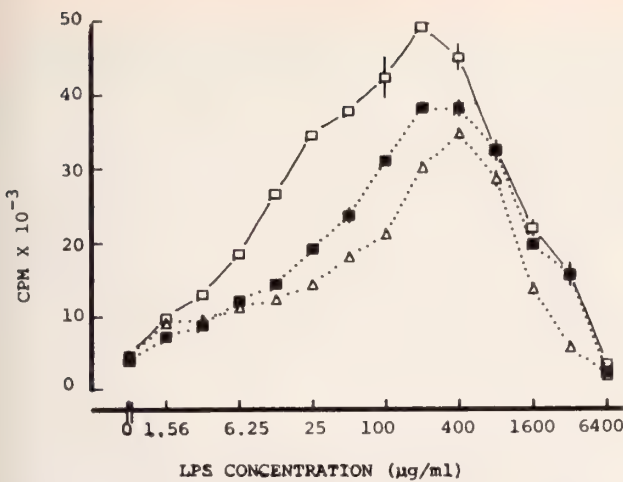


FIG. 12. Inhibition of lipopolysaccharide (LPS) induced responses by seminal plasma fraction A. LPS was from *Escherichia coli* 055:BS phenol extraction method of Westphal. B10.5M mouse spleen cells were stimulated in the indicated LPS concentration. Cells were cultured with LPS alone (□—□) or in the presence of 5 µg (■—■) or 20 µg (△---△) of Fraction A from seminal plasma. Culture conditions and assay procedure were identical to that described in Fig. 9.

cytotoxic to the spleen cells used in the LBT assay in long-term culture of 120 hours. The true inhibition of LPS responses in the dose dependent manner is clearly visible in Figure 12. LPS dose response curve is very broad compared to Con A and we have used the whole range of effective concentration. Inhibition is much more in the ascending than descending part of the peak. Along with the polyclonal activation by FCS data in Figure 8, the data presented in Figure 12 of the inhibition of LPS responses supports the fact that seminal plasma inhibitor depresses B lymphocyte function. LPS is purely a B lymphocyte polyclonal activator in culture conditions employed by us such as serum free media, etc. (48).

e) Inhibition of mixed lymphocyte culture (MLC) reaction by seminal plasma inhibitor is shown in Table VI. MLC is an in vitro test in which the recognition and cellular proliferation is mediated by T lymphocytes and is controlled by H-2 locus (I-region differences) on chromosome 17 in mouse. MLC test has long been considered an in vitro parallel of the allotransplantation reaction in vivo. It represents the recognition phase in which immunocompetent T lymphocyte recognize the foreign alloantigens and are accordingly stimulated to divide. The inhibition of bovine MLC by bovine seminal plasma has been de-

scribed earlier in this paper. Table VI shows that concentration of fraction A as low as 4 µg/ml were able to significantly inhibit an MLC reaction. Inhibition increased with the protein concentration without interfering with the background incorporation in the control cultures except at the highest concentration tested in these experiments. However, since there was no cytotoxicity even at these high concentrations, it has to be assumed that fraction A was also able to inhibit background cell proliferation of normal spleen cells in vitro. These MLC experiments exclude the possibility that the inhibition of Con A induced responses was entirely due to inactivation of the mitogen.

The above studies were conducted under very carefully controlled conditions, for example, utilizing serum-free medium for mitogenic responses and also measuring dose (inhibitor and stimulants) dependent responses.

4. This inhibitor is very potent. For example, as little as 4 µg of a partially purified preparation (and very recently 200–500 ng of a further purified preparation) routinely inhibits the triggering of subpopulations of lymphocytes mediated by Con A in serum-free cultures of 0.5×10^6 spleen cells.

5. This inhibitor is a highly acidic protein with a low PI (Ca 3) and the inhibitory activity is associated with a molecule (or aggregate) of above 100,000

TABLE VI
Inhibition of One-way Mixed Leukocyte Culture Reactions by Fraction A*

Strain of mouse cells in culture	Fraction A (µg/ml)	Cpm/culture
A	—	6,809 ± 387
	4	5,999 ± 409
	20	5,938 ± 443
	100	2,829 ± 271
(A × 5M)F ₁	—	7,832 ± 1,343
	4	10,218 ± 676
	20	7,560 ± 401
	100	2,121 ± 50
A + (A × 5M)F ₁	—	23,628 ± 2,097
	4	17,913 ± 471
	20	16,280 ± 1,579
	100	8,036 ± 389

* Fraction A is the exclusion peak from Sephadex G-100 chromatography of bovine seminal plasma. Spleen cells were cultured for 5 days in serum-free medium (10^7 cells/culture in 1-ml aliquots) in the presence of indicated concentrations of fraction A. The mixed cultures contained 5×10^6 cells of both responder and stimulator cells. The cultures were given a pulse of tritiated thymidine (1 µCi) at between 96 and 120 hr.

daltons (this explains its strong adsorption on spermatozoa which have been shown to have both strongly positive and negative charges).

There is evidence that the suppressive activity is related to a seminal specific protein. Prostaglandins and polyamines found in the semen do not seem to account for the potent immunosuppressive properties described by us and confirmed by others. The active moiety is above 100,000 daltons at least whereas prostaglandins and polyamines are below 1,000 in their molecular weight. On a quantitative level, the inhibition of *in vitro* T lymphocyte responses by seminal plasma is much stronger than that which can be deduced from the normal levels of polyamines and prostaglandins in the semen (E-145, A-40, B-21 and F-6 $\mu\text{g/ml}$). Further, the stability of the suppressive activity in relation to heat sensitivity excludes prostaglandins. Byrd et al (13) have elegantly demonstrated that polyamines require bovine sera for *in vitro* suppression of mitogens, allogenic responses and induction of cell mediated cytotoxicity. Only in our experiments, serum-free cultures were used in mitogen induced responses. Therefore, we could conclusively exclude polyamines.

Confirmation by Others

Stites and Erickson (69) have provided partial confirmation of our hypothesis. The suppressive effect of seminal plasma on lymphocyte activation by phytohemagglutinin and allogenic stimulation was demonstrated. Some of this is subject to criticism. The inhibitory and lymphocytotoxicity activity has been found by us to coexist in the seminal plasma. This was not taken into account by establishing that the seminal plasma at the dilutions tested do not kill the lymphocytes nonspecifically in the control. This may, in fact, explain the strange-dose independence of the inhibitory effects.

Lord et al (37) have provided further support to our findings of inhibitory effects of seminal plasma on lymphocyte responses. The immunosuppressive activity of human seminal plasma was in high molecular weight range (approximately above 200,000 daltons) and the activity was not species specific. It inhibited the blast transformation of various mitogens and allogenic stimuli. They further extended the immunologic characterization by establishing that suppression of the induction phase but not the expression of cell mediated cytotoxicity is affected by seminal plasma.

Levis et al (35) also noted that seminal plasma

was inhibitory to both allogenic and autologous leukocytes stimulated with tetanus toxoid in man. T-lymphocyte are the target of tetanus toxoid mediated responses (17).

Pitout and Jordaan (50) indicated that a dialyzable fraction of seminal plasma inhibits Phytohemagglutinin induced lymphocyte responses. We, as well as others, have found the absence of suppressive activity in the low molecular weight substances of seminal plasma. From the description of the methodology and results described in their paper, the conclusions drawn do not seem to be valid.

Marcus et al (39, 40) have also provided confirmation of our work on immune suppression by seminal plasma in T lymphocyte proliferation by polyclonal activators and allogenic stimuli in mixed lymphocyte cultures systems.

Tarter and Anderson (70) studied the origin of seminal plasma immune response inhibitor in the male accessory glands of mouse. From their results of Con A stimulated culture, and a certain hemolytic system, prostate appears a likely source of origin of seminal plasma immune response inhibitor.

Relationship to Immune Infertility

Infertility in general is considered a curse. The cause of reproductive failure has been the subject of intensive research over several decades. What resulted from these studies to date is briefly summarized.

About 2%–10% of couples seeking medical advice for their infertility problems are believed to be infertile from immunologic causes. Infertility caused by immune responses to seminal antigens in male and female have been recently reviewed (6, 7, 64). The routinely used techniques are exclusion of other abnormalities and demonstration of circulating antibodies by spermagglutination, sperm immobilization, spermatotoxicity and immunofluorescence techniques. Some of the patients have been treated by condom therapy to avoid the contact of seminal antigens with female body (6) or immunosuppressive drugs (21, 65) or artificial insemination (66).

All these procedures have come under close scrutiny. Until immunologic systems impairing fertility and techniques for assessing alleged immune reactions in fertile subjects are carefully defined, the entire field of immunoreproduction will, according to Katsh et al (33), "continue to languish in speculative limbo." The presence of humoral immunity to seminal components has not been definitely

found as a close correlation with infertility (31). Some fertile individuals had been demonstrated to have circulating antibodies. There has been a constant debate on the diagnostic procedures used. As a point of fact, a significant proportion of the published work in this area suffers from the lack or required understanding of immunologic diagnostic procedures, or special pitfalls of this technology when applied to sperm or sometime unavailability of proper facilities in the diagnostic infertility clinical laboratories. All this has clouded the conclusions and muddled the perspective resulting in a somewhat discouraging note on the progress in this field. Also, from theoretical considerations, cell mediated immunity could be important since seminal antigens have not been considered thymus independent. Methodology for the study of cellular immunity in this area has not yet been developed. This has led people to wonder whether we do have or do not have proper diagnostic methods for detecting immune infertility. A reasonable answer would be that currently available methods are good indicators when properly used but a considerable improvement can be made in their efficacy in the near future. It is difficult to evaluate the usefulness of various methods of treatments at the present time because of the uncertainty that they were directed to the cause of true immune infertility in the first place for comparison to be valid.

Since the science of immunology has made great progress in the last decade, systematic study of immune infertility rather than the immediate clinical problems is likely to provide small but definite increment to the understanding of this evasive complex problem.

The precise quantitation of immune response and the effector mechanism by which either cellular or humoral immunity brings about infertility are poorly understood. The so-called "immune factors" in immune infertility are IgM, IgG or IgA type of antibody. The mediators elaborated by sensitized T lymphocytes in immune infertile patients have not been identified so far. Also, cytotoxic T lymphocyte may constitute another class of "immune factors." Another way of looking at "immune factors" is by the techniques of detection such as agglutinating, cytotoxic, immobilizing type of antibody, or cell mediated immunity (CMI) factors.

Methodology for the study of cellular immunity in this area has not been developed. Blast transformation of lymphocytes from male and female patients by spermatozoa has been used in unexplained infertility (25, 28, 72). This method is unlikely to work since a potent "immune response inhibitor"

in seminal plasma has been identified by us (54, 57) and confirmed by others (50, 37, 39) and it is difficult to remove all adsorbed seminal plasma from spermatozoa by washing (49). Delayed hypersensitivity has been elegantly demonstrated against a glycoprotein component of seminal plasma by Halpern et al (26a) and confirmed by others (15, 67). Assays for migration inhibition factor (MIF) and leukocyte adherence inhibition (LAI) were attempted (71, 60) in mouse system but appear far from satisfactory for use as a diagnostic test for human infertility at the present time.

The immune response provoked in the female as a consequence of coitus or in the male as a consequence of leakage of sperm autoantigens has not been shown to be relevant to infertility. The relevant "infertility sperm antigen" remains undefined at the present time.

Antibody is not sufficient to neutralize sperm function. The presently known assays for humoral immunity measure *in vitro* agglutination or reduction of motility or death of the cell in spermatotoxicity. Antibody bound to spermatozoa can also be detected by immunofluorescence. But, in all these four types of tests, 100% of the spermatozoa are never involved. The results of artificial insemination from frozen bull semen all over the world involving million of cattle clearly establish that 1/1000 dilution of ejaculate routinely give rise to over 60% chances of fertilization with one single insemination. Certainly out of 250 million human spermatozoa deposited during a single coitus, enough are left free to succeed in penetration of egg in spite of a strong "immune factor." Also, studies on the transport of sperm in the female reproductive tract have clearly established that muscular upward contractions of the female reproductive tract rather than intrinsic motility of spermatozoa is important to account for rapid movement of spermatozoa from the site of deposition to the site of fertilization immediately after coitus.

The action of immune factors on spermatozoa alone can not adequately account for immune infertility. Another hypothesis is that spermatozoa deposited during coitus act as immunogen but the action of "immune factors" is on blastocyst. This hypothesis looks very attractive on the basis of the following facts.

- 1) Antigens on spermatozoa and blastocyst of several mammalian species share common specificity which may be controlled by some primitive genes conserved during evolutionary development. Francois Jacob (30, 11) and his collaborators at Pasteur Institute, using a specific antiserum against

F9 cell line developed from mouse teratocarcinoma, have established cross reaction between mouse spermatozoa, human spermatozoa, and mouse cleavage embryos employing cytotoxicity, immunofluorescence and absorption techniques.

The cross reaction between sperm and blastocyst was also pointed by Alan Menge and his collaborators of the University of Michigan as early as 1974 (44) when they observed that uterine fluids of rabbits immunized with spermatozoa were embryotoxic and this effect could be abolished by absorption of the immune SIgA fraction with spermatozoa. More recently, they have demonstrated the presence of some of the mouse sperm antigens on ova and early embryos using indirect immunofluorescence technique (45).

2) Some immune infertile women are spontaneous early repeat aborters. Warren Jones (31) at Flinders University of South Australia studied 195 infertile couples consisting of 106 primary unexplained, 64 secondary unexplained and 25 with minor organic causes of infertility and followed them for abortion while testing for the presence of anti-sperm antibody. Table VII shows the results of this study.

It is evident that females positive for sperm antibody have a high spontaneous rate of abortion compared to the negative controls and the difference is highly significant statistically. With the β -HCG test, very early pregnancy and abortion can be established so that antisperm antibody can be related more reliably with spontaneous early abortion.

3) From the view point of the efficiency of "immune factors" to cause infertility, it appears more logical that a 16 cell blastocyte is a more appropriate target than 250 million spermatozoa deposited during a single coitus.

If, in reality, blastocyst is the target of antisperm antibody, it is easy to visualize the reason for confusion surrounding the value of antibody detection methods as well as the success with the treatment protocols.

Immunologic tests are sensitive tests and it is easy to obtain false positive results. In most of the clinical diagnostic laboratories, a known negative and a known positive control are not run routinely, adding to the unreliability of reported results.

The pitfalls of each of the four routinely used diagnostic methods are as follows. Spermagglutination is most widely used and one wonders whether it is specific or nonspecific for antibody. Unfortunately, spermatozoa are prone to natural

agglutination under a variety of conditions in the absence of specific antibody. Human spermatozoa has been reported to undergo agglutination when exposed to some microorganisms, viruses, certain chemicals, and other factors present in fresh autologous serum. Boettcher (9) ascribed sperm agglutination in infertile patients to steroid and beta lipoprotein like protein complex. Ingerslev (29) had contradicted the steroid protein complex concept but agrees that spermagglutinins are not antibody. Because of the apparent unreliability of agglutination test, it may be desirable to abandon it. Sperm immobilization test has two difficult requirements that must be met to give reliable results. The sperm specimen must be highly motile and the test manipulations including microscopic viewing should be conducted at 37°C. Spermatotoxicity technique has not been applied in clinical use widely so far, but has tremendous promise in the future.

Immunofluorescence is an excellent technique but has been widely misused for detection of anti-sperm antibody. It is well known that fixation with alcohol increases nonspecific background fluorescence with spermatozoa, yet few workers use unfixed single cell suspensions of spermatozoa in their studies. A high fluorochrome to protein ratio of antisera gives guaranteed nonspecific positive fluorescence, but few authors define their reagents or fail to titrate them to choose the right dilution. Further, if the excitation wavelength is a wide band, specificity decreases considerably and false positivity increases.

In summary, considerable improvement in the diagnostic techniques can be achieved by simple application of the current immunologic knowledge. Three important areas are: 1) substitution of quantitative instead of qualitative parameters in the assays, 2) use of more rigorously controlled and defined immunologic reagents and strict adherence to general immunologic principles, 3) introduction of micromethods that utilize the property of paraffin oil to check the evaporation. Micromethods would permit more detailed analysis of biological fluids with much lesser cost and time and use of the same semen sample in more tests.

To test the hypothesis experimentally that males in true immune infertile marriages have either absence or lower quantitative levels of immune response inhibitor compared to fertile men, it is important to identify true immune-infertility. The present state of art in the diagnosis of immune-infertility is far from satisfactory to reach this objective.

TABLE VII
Spontaneous Abortion Rates in Infertile Couples

Sperm Antibody	No. of Patients	Total Pregnancies	Spontaneous Abortion No.	Abortions %
Immobilization	Negative 177	94	12	12.8%
	Positive 12	12	9	75.0%
Microagglutination	Negative 149	89	11	12.4%
	Positive 6	27	11	40.7%
Both tests	Negative 143	76	8	10.5%
	Positive 12	9	6	66.7%

From Jones, 1977.

Discussion

Our experimental results clearly demonstrate that seminal plasma contains substances that suppress T-lymphocyte responses to specific and non-specific stimuli. Thus, both Con A-induced and mixed lymphocyte responses, known to be carried out by T cells (47, 48), were consistently inhibited by a fraction of seminal plasma that displayed no cytotoxic activity on cultured lymphocytes. The inhibition of the T-cell-dependent in vitro antibody response to sheep erythrocytes also suggests T-cell suppression as the mechanism of inhibition. However, since the same seminal plasma fraction was also found to inhibit the direct (T-cell-independent) B-cell activation caused by FCS, it appears more likely that the B cells are also targets for the inhibitory activity of seminal plasma. The same fraction described here is able to inhibit the lymphocyte proliferative responses to lipopolysaccharide, which is a selective B-cell mitogen (48). This substance has met the most rigorous criterion of true inhibition of immune response and in fact, appears very potent in its activity. It has been tested successfully by such desirable features of the assay system as dose-dependent inhibition, use of effective range of mitogen, cytotoxicity control over long periods of cultures, use of serum free media for mitogen response.

The suppressive influence of the seminal plasma components could be expressed at the level of cell activity, for example, by preventing activation by the competent mitogen or antigen, or at the level of expression of the activated phenotype, for example, by preventing the activated cells from proliferating. Studies are in progress to answer these questions, as are investigations of the effects of seminal plasma inhibitors on cell types other than T cells, such as B cells and macrophages and subpopulation of T and B cells. Preliminary results exclude the possibility of macrophage as target for the immune response inhibitor.

The experimental results support earlier speculations on the physiologic relevance of the immunosuppressive capacities of seminal plasma (53, 54, 57). Thus, since the female genital tract is susceptible to immunization (5, 18, 63), why do normal adult females not respond to the male sperm and seminal plasma antigens? It has been demonstrated previously that male sperm is actively phagocytosed and transported to the regional lymph nodes after deposition in the vagina (59); nevertheless, no immune response develops. On the other hand, it is well known that ejaculated spermatozoa display tightly adsorbed seminal plasma proteins on the surface. Therefore, it appears likely that the female immune system is simultaneously presented with antigenic stimuli and with the inhibitor proteins in the seminal plasma that we have described in the present paper.

Actually, there is direct evidence to support the assumption that the seminal plasma inhibitor is responsible for the immune unresponsiveness to ejaculated sperm, since epididymal sperm, which lacks seminal plasma proteins on the surface, is able to induce an immune response once it is deposited in the female reproductive tract (5). The importance of these observations rests on the fact that a female immune response to the male sperm sometime results in sterility; they thus open an entirely new approach to problems of immune infertility and immunologic regulation of fertility for contraception.

Conclusion

Semen of mammalian species contains a built-in system to prevent immunologic sensitization of females against antigens of sperm and seminal plasma in spite of repeated coitus. This prevention of sensitization is due to the presence of a potent immune response inhibitor originating from one of the sex accessory glands of the male. The inhibitor has been biologically characterized so far.

Failure of this built-in system in the male results in immune response to seminal antigens in the female. This seminal immunity when, of the right kind and magnitude, results in clinical immune-infertility.

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Common Denominators of Pregnancy and Malignancy

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Abstract

Antigenic tissue is paradoxically tolerated by a seemingly intact immune system in two biological situations, pregnancy and malignancy.

Increasing evidence in recent years supports the idea that similar or identical immune mechanisms may be responsible for the tolerance of antigenic tissue in these two systems. What is apparently essential for the immunologic survival of the fetal allograft is detrimental for the patient with malignancy. This evidence is based on a number of common denominators, including such general observations as depression of cellular immunity, the presence of suppressive and/or blocking substances, embryogenization of neoplastic tissue, common lymphocyte populations and activities, and some specific in vitro observations, such as leukocyte migration enhancement (LME) and red cell immune adherence (RCIA).

These common denominators of pregnancy and malignancy are reviewed in detail and we suggest that further understanding of one system will also contribute to our knowledge of the other.

Introduction

Pregnancy and malignancy are antithetical events in the human life cycle, the first representing the regeneration of life, and the second frequently portending death. They may, however, represent differing clinical situations based on the same immunologic mechanisms. Recent evidence indicates that the immunologic mechanisms that are of benefit to the survival of the fetal allograft within the maternal immune system may be detrimental to the survival of the cancer host. Much of this evidence is circumstantial; however, the picture becomes more convincing as many of the immunologic mechanisms identified in malignancy are also found in pregnancy.

Immunologic research has so far been undertaken mainly by investigators in the field of oncology. Similar research into the immunologic situation of pregnancy has lacked motivation and emphasis. Recently, however, the recognition has grown that knowledge in either of the two fields may benefit

the other. Thus, there has been more emphasis on research in the field of pregnancy immunology, and the subsequent identification of additional common denominators of these two unique immunologic conditions.

In this review, we will try to summarize all the currently recognized common denominators of pregnancy and malignancy in an effort to identify the immunologic mechanisms that may be responsible for the tolerance of antigenic tissue in either situation.

Common Denominators of Pregnancy and Malignancy

These common denominators may encompass a wide variety of clinical, biochemical, pathologic, and purely investigative in vivo or in vitro observations. In this summary, we will make an arbitrary subdivision of factors, but this should not be taken as indicating a clearly defined separation of individual determinants. Moreover, we will stress the interdependence among many of these factors throughout the discussion of possible active mechanisms.

It should also be recognized that common denominators represent no more than hypotheses at the present time. Further investigations, assuming a common mechanism, will be essential before these

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common denominators can be termed common factors.

Depression of Cellular Immunity

Pregnancy is generally considered to be a state of depressed cellular immunity. This theory is supported by clinical manifestations such as an increased incidence and severity of infections (1, 2) and enhancement of malignant tumors (3-5), in vivo observations, such as prolonged persistence of human skin grafts during the third trimester of pregnancy (6), and depressed skin reactivity to tuberculin during pregnancy (7) and in vitro assays, such as depressed maternal lymphocyte stimulation with mitogens like phytohemagglutinin (PHA) (8, 9). Animal models confirm a nonspecific reduction of cell-mediated immunity, for instance during the second half of pregnancies resulting from matings between H-2 incompatible mice (10).

Similarly, malignancy is associated with a decline in cell-mediated immune potential. In reproductive malignancies, immunologic responses in general were shown to correspond to a clinical stage: the more advanced the malignancy in an individual patient, the more depressed cellular immunity was found to be. The assay systems used were similar or identical to those noted in association with pregnancy (11-15).

Thus, both pregnancy and malignancy are associated with decreased cellular immunity. However, the implications of this finding are still unknown. It has not been determined whether a decline in cellular immunity does in fact predispose a patient to the occurrence of malignancy, or whether this decline permits the acceptance of the graft. Whatever the immunologic mechanisms resulting in tolerance of malignancy and pregnancy, it is also conceivable that they act independently of those mechanisms causing a decline in cellular immune capacity. The decline in cellular immunity may thus represent an immunologic situation occurring secondary to a still undefined, primary event.

The evaluation of cellular immune capacity by any of the methods noted above is still difficult and rarely satisfactorily reproducible. Cell-mediated immunity is dependent on a number of widely varying factors. Surgery, for instance, has repeatedly been shown to affect immune competence (16,17), and many of the patients involved in immunologic studies of cancer and/or pregnancy populations have undergone surgery shortly before their evaluation. Similarly, factors such as age, sex, and intercurrent disease are often inadequately controlled for, despite their clearly established significance (18).

In summary, decreased cellular immunity does represent a common denominator of pregnancy and malignancy. However, more sophisticated investigations of the underlying mechanisms and of the assay systems utilized could render these findings more meaningful. For example, cancer-specific density changes in lymphocytes after stimulation with PHA were recently reported (19). If similar changes could be demonstrated in pregnancy lymphocytes, it would reinforce the theory of decreased PHA stimulation common to both systems.

Suppressive (Blocking) Substances

Substances that suppress and/or block cellular immune reactivity during pregnancy have been reported by a large number of investigators. Table I lists a variety of such substances by their biochemical characteristics or mode of action, as identified by in vitro laboratory investigations of pregnancy.

1. Immunoglobulins and circulating immune complexes: The earliest observations of maternal

TABLE I
Substances that have been Implicated in Suppressing (Blocking) the Immune Response and the Assay Systems Applied (During Pregnancy)

Substances	Assay Systems	References
Serum*	Colony inhibition test (mouse)	20
Plasma*	Mixed lymphocyte reaction (human)	22-24
Serum*	PHA-stimulation test (human)	25
Plasma*	Macrophage migration inhibition test (human)	26
Serum*	Polymorphonuclear leukocyte phagocytosis (human)	27, 28
Plasma (IgG)	Macrophage migration inhibition test (human)	29, 30
Serum (IgG)	Microcytotoxicity assay (mouse)	31
Immune complexes	Various (see reference)	32
Placental glycoprotein	Lymphocyte transformation (human)	33
α -globulin	PHA-stimulation test (human)	34
Pregnancy zone protein	PHA-stimulation test (human)	35
Hormones	Nonpregnant experience**	
Estrogen	Various assay systems	36-41
Progesteron	Mixed lymphocyte reaction (human)	45
HCG	Various assay systems	46-48

* Undefined component

** In vitro experience that may be extrapolated to pregnancy because of high circulating hormone levels.

blocking substances in pregnancy sera were made by the Hellströms and coworkers. As early as 1969, they noted that cellular immunity to antigenically foreign mouse embryonic cells could be abrogated by a serum factor (20). Interestingly, these pregnancy investigations were inspired by earlier studies in tumor immunology that had demonstrated antibodies able to block the lymphocyte effect against tumor cells (21). A common denominator of the two fields was thus established at the early stages of investigation.

In earlier investigations of both plasma and serum of pregnancy, general blocking (suppressing) activity was demonstrated, without attempting biochemical identification of the blocking substances (22–28). Recently, it has been claimed that such activity could be discerned in plasma only, and not in serum (24).

Biochemical investigation made it apparent that a large number of substances present in pregnancy plasma and/or sera had immunodepressive potential. The Hellströms and coworkers demonstrated that the blocking substance they had described earlier in sera of multiparous mice actually contained immunoglobulins of the IgG class (31). Rocklin's group was able to confirm the involvement of an IgG immunoglobulin in the suppression of maternal responsiveness to paternal antigens (29). Later the same group of investigators confirmed their findings by demonstrating that habitual aborters frequently lack this specific IgG fraction, indicating that the presence of this fraction is essential to the survival of the fetal allograft (30).

In further defining the blocking immunoglobulin fraction, the Hellströms and associates concluded that IgG_{2a} and IgG_{2b} form a complex with embryonic antigens in the mouse when exerting their blocking influence (31). This discovery represented another common denominator with malignancy because it indicated the presence of immune complexes in pregnancy, as had been observed previously by the same, and other, investigators in malignancy (46,47).

The presence of immune complexes in the maternal circulation during normal and abnormal pregnancy is still a controversial issue (32). However, if the presence of circulating immune complexes in pregnancy were confirmed, the field of pregnancy immunology would be opened to a variety of exciting possibilities. The influence of immune complexes on the immune system could be implicated in any of a variety of functions (32), similar to other very specific immunologic situations such as neoplasia and autoimmune disease.

In pregnancy the immunoactive IgG fraction was shown to be HLA-D dependent (30,48,49). This observation is important because HLA-A-, B-, or C-determined antibodies have been associated with graft rejection (50), whereas HLA-D-determined antibodies (which are cytotoxic to B lymphocytes (49)), have been correlated with graft enhancement (51).

Thus, considerable evidence suggests that in both cancer and pregnancy an IgG immunoglobulin may participate in changes in the immune system that permit the tolerance of antigenic tissue. The presence of Fc-receptors on animal and human placenta (52, 53), and on human malignant tissue (54, 55), represents indirect evidence that immunoglobulins may be involved in immunologic mechanisms specific to both conditions.

2. Other substances: Substances that do not contain antibody were implicated as well. These include a number of placental proteins (33), circulating globulins, mainly of the α -fraction (34), and a seemingly endless number of so-called pregnancy zone proteins (35). The latter group is especially difficult to define as newly discovered fractions are reported almost daily. Not all of these fractions contain immunosuppressive properties (56); however, it is interesting to note that some of these proteins apparently occur not only during pregnancy but also in choriocarcinoma and other neoplasias (57). Thus, another common denominator between the two systems may be established.

The importance of the immunosuppressive effect of various hormones is even more difficult to establish. Considerable evidence has accumulated indicating that almost all hormones suppress immune activity at certain concentrations. During pregnancy, as opposed to the nonpregnant state, many hormone levels are elevated. However, their role in the survival of the fetal allograft has remained dubious. There is no doubt that the three important hormones of pregnancy, estrogen (35–41), progesterone (42), and HCG (46–48), possess immunosuppressive potential. None, however, is more clearly dose dependent than HCG (46, 47, 58); and none is as clearly associated with malignancy as HCG. We will discuss the implications of the presence of HCG in both pregnancy and malignancy in greater detail in the following section.

Embryogenization

In an earlier article reviewing this topic, we coined the term "embryogenization" to describe several tumor-associated phenomena that are limited to the embryonic state of development and

malignancy (59). They are characterized by the following three determinants:

1. *Dedifferentiation*: When the term embryogenization is applied to malignancy, the first correlation is always made to the histologic differentiation of the specimen. The stage of differentiation of many tumors is closely related to the prognosis of the patient (61); anaplasia of cells represents a primitive state in their evolution (60). In fact, neoplasia has been considered by some researchers to represent a disease of aberrant differentiation. One proponent of this hypothesis, Prehn, suggests that neoplasia occurs as an aberrant and abortive attempt to form a regenerative blastema. He cites as one of the most instructive examples of redifferentiation the occurrence of so called paraendocrine syndromes, in which neoplasia produces one or more hormones that would be produced in normal situations by some unrelated organ (62). In general, the most undifferentiated tumor in an organ will acquire the "embryogenic omnipotence" to secrete hormones (63). Thus, the histologic dedifferentiation to an earlier embryogenic state of malignancy represents another common denominator with pregnancy.

2. *Paraendocrine syndromes (including HCG)*: As noted, a variety of hormones may be secreted by inappropriate tissue once the tissue has become malignant.

One of the frequently produced ectopic hormones in both gynecologic (64, 65) and nongynecologic (66, 67) neoplasias is parathyroid hormone. However, the hormone that seems to be most relevant to our topic is HCG. Its function in the preservation of pregnancy, especially during the early stages, is well recognized. Placental function does not require any comment here other than a reiteration of its importance as an endocrine factory that takes over from the corpus luteum, and a mention of its possible immunologic influence, which has not been considered quite so extensively.

We noted earlier that HCG, like other hormones, is capable of immune depressive activity in certain concentrations (46-48, 58). It is therefore of special interest that this hormone is present not only during pregnancy, but has also recently been recognized, with the help of more sophisticated assay systems, on the surface of many, if not all, neoplasias (68-72). The latest reports indicate that minute amounts of HCG or HCG-like material may even be present on normal tissue, and may be associated with rapid cell proliferation (73, 74).

The special ability of HCG to adhere to the surface of malignant and trophoblast cells (68) may imply a possible immune function. Its adherence to the surface of cells may cause changes in tissue

antigenicity. These changes may either render the tissue nonantigenic by covering antigenic sites and thus preventing them from exposure to immunocytes, or they may create a mechanism by which enhancing substances are produced (59).

To date, ectopic hormone production by neoplastic tissue is imperfectly understood. A number of hypotheses have been advanced in the literature, but none has yet provided a satisfactory explanation. In one interesting recent study, a protein of molecular weight 65,000 was found to be the translation product in all cases in which messenger RNA was extracted from various hormone-secreting tumors and injected into *Xenopus* eggs. This study supports the hypothesis that a primary gene product of all different types of carcinoma cells is a single large hormone precursor containing different specificities. A specific enzyme system in different tissues selects specific hormones that are then liberated from the larger primary protein (75). This observation, and the following suggestions, may represent an explanation for the detection of partially biologically inactive HCG (73), and hence a tentative link between the secretion of hormones and oncofetal antigens by tumor cells.

3. *Oncofetal (embryogenic) antigens*: The discovery of oncofetal antigens in a variety of malignancies was greeted with great excitement in the early 1960s. At first, some of these substances were believed to be available possibly for diagnostic as well as monitoring purposes in malignancy. Unfortunately, later investigations showed that none of the applied assay systems was adequately specific to either function since some of these antigens are also present in nonmalignant conditions.

Carcinoembryogenic antigen (CEA) is a good example of the ups and downs in research experience with oncofetal antigens. Since its discovery in the gastrointestinal tract by Gold and Freeman (76), CEA has been found in a variety of human malignancies (77-84). Other oncofetal antigens that have been found in association with neoplasia are α -fetoprotein (AFP) and, less frequently, pancreatic embryonic antigen (PEA), to mention only two (77, 85-91). Some tumors may produce more than one of the oncofetal substances, which may support the theory mentioned above of a common precursor molecule (75). Thus, CEA and AFP have been reported to be elevated in some cancers (78), while in others HCG and AFP were concomitantly elevated (92, 93). This latter observation has provided further support for postulating a link between the production of oncofetal antigens and HCG in tumor patients.

Earlier, we mentioned that the Hellströms were

able to demonstrate, in their original experiments on multiparous mice, that immune complexes containing embryogenic antigens complexed with IgG immunoglobulins were in fact the immunologic blockers in serum (31). It was thought that perhaps a similar embryogenic antigen-containing complex might be involved in blocking activity in cancer (59). In general, it was assumed that oncofetal antigens, such as CEA, are not involved in the stimulation of cell-mediated immunity against neoplasia in the human. However, even at that time a specific antibody-immune response against neoplasia had been recognized with some of the fetal antigens (79). More recent data from a variety of animal experiments indicate that fetal antigens are in fact capable of producing a very significant immune response on a number of different levels (94-97). In the human experience, a significant IgM anti-CEA antibody response has been detected in patients with digestive system cancers that have not metastasized. In cases with proven metastatic spread, this response was not detectable, indicating the formation of antigen-antibody complexes when there has been metastasis (79). The importance of these findings for the eventual explanation of tolerance mechanisms involving oncofetal antigens will be discussed later. It should be noted here, however, that CEA levels in malignancy are generally higher in the presence of disseminated disease (79-84), indicating that their production is dependent on tissue mass. CEA-anti CEA antibody immune complexes exist in cancer patients (98) and have also been shown in the circulation in both pregnancy and malignancy (99). It is tempting to assume that the presence of oncofetal antigen-antibody complexes in both situations may play an important role in the induction of immunologic tolerance. Similar thoughts have been entertained by a number of investigators (100-103).

Lymphocyte Populations

It would exceed the scope of this review to discuss lymphocyte subpopulations in detail. However, a discussion of the common aspects of lymphocyte immune activity in pregnancy and malignancy is quite relevant to our topic. It has been recognized for some time that, in the tumor system, the T lymphocyte (in any of its varied subgroups) does represent an effector cell in the cellular arm of the immune system, with different antigens being destroyed by different T cell subgroups (104). The interaction of different T cells in the tumor model has been described in detail (105). This degree of sophistication has not yet been achieved in the

immunologic research of pregnancy; however, some points of interest have been targeted. For instance, the early identification of suppressor T cells in patients with malignancies led to similar investigations of pregnancy. Early research in the human model failed to demonstrate suppressor cell activity in normal pregnancy (59), but more recent studies with a number of animal models have yielded data with conflicting results (Gottesman SRS and Chaouat G, in separate presentations, The First S. B. Gusberg Seminar on Reproductive Immunology, New York, June 27, 1980). Further investigation is, therefore, clearly indicated.

Suppressor cells, however, have been established beyond any reasonable doubt in fetal cord blood (106, 107). These suppressor cells are probably activated transplacentally, raising questions as to the maternal impulses responsible for activation. Oldstone and associates suggested that immune complexes may in fact activate these cell populations (106). Fetal suppressor lymphocytes were shown to suppress not only fetal lymphocytes, but also maternal cells. It can therefore be postulated that this fetal suppressor capability plays a role in the immunologic adaptation of the maternal immune system to pregnancy. Even in the absence of proven maternal suppressor cells in pregnancy, a common denominator can be hypothesized with data from the fetal compartment.

Common in vitro Observations

One of the major emphases in our laboratory over the past few years has been the identification of common denominators between pregnancy and malignancy on the assay level. Our interest was based on the theoretical background that has been delineated in the previous sections. However, the first in vitro phenomenon that we recognized as being specific to both pregnancy and malignancy was discovered more or less by accident (108). We have now discovered a common denominator in both the white cell and the red cell lines.

1. *Leukocyte migration enhancement (LME)*: In the mid-1970s, we noticed that incubating pregnancy white blood cells with amniotic fluid in leukocyte migration chambers resulted in a consistent pattern of migration larger than that of either control leukocytes or control antigens. In reference to the well-established phenomenon of leukocyte migration inhibition (LMI), we termed the pattern we observed leukocyte migration enhancement (LME) and assumed that it was caused by a blocking factor in amniotic fluid (109). We then extended our investigation to a larger study population and were

TABLE II

*Leukocyte Migration Enhancement (LME) with Amniotic Fluid and Pregnancy Plasma as Compared to Control Subjects**

	LME with either amniotic fluid or plasma	
	MI $\pm$ SD	MI $\pm$ SD
First trimester	110.0	
Second trimester	133.2	189.4
Third trimester	140.3	139.8
Total	132.5 $\pm$ 41.7	172.9 $\pm$ 73.0
Controls	90.7 $\pm$ 29.0	102.1 $\pm$ 57.4
Significance	p < 0.003	p < 0.03

* Modified from Gleicher N, Beers P, Kerenyi TD, Cohen CJ, Gusberg SB: *Am J Obstet Gynecol* 1980; 136:1-4 (110).

able both to confirm our earlier observations with amniotic fluid and to demonstrate LME with pregnancy plasma (Table II)(110). With LME established as a pregnancy-specific in vitro phenomenon, we intended to turn our attention to the investigation of LME in cancer patients. It was at this stage that one of our male control patients showed the classic migration pattern that had usually been associated with pregnancy. Four days later he was diagnosed as suffering from an osteogenic sarcoma of the left femur. This case led to the investigation of a larger number of patients with different types of malignancies. As expected, LME was observed in cancer patients in a pattern very similar to that of pregnancy. Interestingly, amniotic fluid did not cause LME in cancer patients, as it had done in pregnant patients, but plasma did cause LME in cancer patients even more obviously than it had in pregnant patients. From this investigation we concluded that LME was not only dependent on a factor in either amniotic fluid or plasma, but also on a specific cell population. In other words, specificity of both fluid fraction and cell population was necessary to achieve LME in vitro (111). Our findings in cancer patients are demonstrated in Table III.

In an attempt to investigate the mechanism of LME in greater detail, we then demonstrated that the phenomenon was reproducible not only with the leukocyte migration assay, but also with the indirect migration inhibitory factor (MIF) assay. LME was caused by antibody excess and could be transformed into LMI by adding antigen until antigen excess was reached. Finally, the amniotic fluid and plasma fractions involved were within the IgG immunoglobulin fractions, and by selectively removing IgG, LME could be converted into LMI (112).

Most importantly, LME was established as an in vitro common denominator of pregnancy and ma-

lignancy. Unfortunately, leukocyte migration inhibition is difficult to quantitate. Our laboratory investigations continued, therefore, in an attempt to find another in vitro assay that would be more effectively quantitative and still be specific to both conditions. This in vitro system would be immunologically applicable and represent the in vivo "happening" of immunologic tolerance. Such a system has recently been developed in our laboratory.

2. *Red cell immune adherence (RCIA)*: We have recently developed an assay system that allows us to evaluate the relative red cell immune adherence (RCIA) receptor activity of individual patients. This assay is based on antibody-coated sheep red cells that adhere to human red cells in the presence of complement and thus form mixed human-sheep red cell aggregates (RR-rosettes). The quantitative evaluation of RCIA activity is made possible by calculating the percentage of human red cells adhering to one or more sheep red cells (113). Preliminary investigations in immunologically unique situations, such as neoplasia and autoimmune disease, revealed statistically significant differences in RCIA activity among different groups (114). These observations led us to postulate a very precise function of the red cell immunocyte within a red cell dependent immune system (115).

RCIA was established as a common denominator of pregnancy and malignancy because both conditions are characterized by decreased RCIA activity (114, 116). This is an especially important common denominator because we were also able to define a patient population that showed the opposite phenomenon, that is, increased RCIA activity. This group of patients included various autoimmune states (114).

It remains to be seen how specific a role RCIA activity will play in the evaluation of individual patients with different medical disorders. In pregnancy, one may assume that there are aberrations from the normal pregnancy picture in RCIA activity in situations where there is malfunctioning of the normal immune adaptation of the maternal host to

TABLE III

*Leukocyte Migration Enhancement (LME) with Amniotic Fluid and Cancer Plasma as Compared to Control Subjects**

	LME with either amniotic fluid or cancer plasma	
	MI $\pm$ SD	MI $\pm$ SD
Cancer Patients	105.0 $\pm$ 50.6	180.7 $\pm$ 86.4
Controls	90.7 $\pm$ 29.0	102.1 $\pm$ 57.4
Significance	Not significant	p < 0.04

* Modified from Gleicher N, Beers P, Cohen CJ, Kerenyi TD, Gusberg SB: *Am J Obstet Gynecol* 1980; 136:126-131 (111).

pregnancy. Such situations may best be represented by the syndrome of EPH-gestosis or habitual abortions.

Concept of Tolerance

The development of tolerance to antigenic tissue probably represents the most important among the many poorly understood mechanisms in human reproduction. It encompasses not only the ability of the maternal host to tolerate the fetal allograft but also the ability of the fetus to tolerate its own as well as maternal antigens.

The importance of understanding the mechanism(s) of tolerance does not rest with its reproductive repercussions, but is even greater in its application to the understanding, therapy, and cure of disease. In other words, the same mechanism that allows survival of the fetal allograft within the maternal immune system may permit malignant tissue to survive within the cancer host. Thus, what is beneficial in pregnancy may be detrimental for the cancer patient. Understanding the mechanism of fetal allograft survival may thus benefit not only our understanding of pregnancy-associated phenomena, but also our understanding of cancer biology, transplantation biology, autoimmune disease, and a large number of related and still poorly understood clinical phenomena.

It would exceed the scope of this survey to review all the published theories and suggested mechanisms relating to the development of tolerance. One concept, however, deserves some additional commentary in the framework of this article. In a previous publication we suggested the existence of a mechanism of tolerance induction that was dependent on the presence of oncofetal (embryogenic) antigens (59). In our opinion, this is the only concept yet proposed that comprises tolerance induction in the systems of both pregnancy and malignancy.

According to this concept, which has been expressed similarly by a number of investigators (100-103), oncofetal antigens are essential for the induction of tolerance. Proof, however, is still lacking. The attractiveness of this theory lies in its apparently simple explanation for the redepression of oncofetal antigen genes (and possibly oncofetal endocrine genes) in neoplasia. Although experimental proof for such a theory should not be too difficult to obtain, little data has yet been gathered.

In describing some common denominators of pregnancy and malignancy, we hope to suggest some research directions that may be taken in this important field of immunology. Whether the prob-

lems we have outlined here are approached from the point of view of neoplasia or pregnancy, we believe that any new knowledge from either field will ultimately benefit both.

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Trophoblastic Disease, A Bridge Between Pregnancy and Malignancy

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Abstract

The morphology and biological behavior of the early trophoblast is reviewed. Light and electron microscopy studies of cyto- and syncytiotrophoblast offer some insight into the immunologic processes of nonrejection and self-limited growth of the trophoblastic tissue.

Trophoblastic diseases: spontaneous abortion, transition mole, hydatidiform mole, invasive mole (chorioadenoma destruens), and choriocarcinoma are reviewed and their pathogenesis is discussed in relationship with the failure of the maternal defense mechanism and the aggressive growth of the trophoblast. Local (hydatidiform and invasive moles) and systemic (choriocarcinoma) invasion by the trophoblastic tissue, may have a more or less favorable response to chemotherapy if correlated with specific morphologic features of the tumor and of the host response.

The trophoblastic gestational tumors originate in the normally invasive growing trophoblast. The factors able to transform a physiologic growth process in an immunologically modified host, into a neoplastic growth in an immunologically deprived host may constitute the bridge between pregnancy and malignancy, the understanding of which may shed some light on carcinogenesis in general.

Trophoblast, Morphology and Function

"Trophos" means nutrition in Greek. Nourishing the developing fetus represents one of the basic functions of the trophoblast. In order to fulfill it, and to be able to transfer substances between fetal and maternal organisms, the trophoblast has to grow and develop, from the primitive outer layer of the blastocyst, to a vastly ramified and complex structure covering the placenta.

The growth pattern of the trophoblast during the process of implantation in normal gestation represents a unique, though poorly understood, biological process. It represents the growth of one individual entity into another. The growing tissue differs from its host both genetically and antigenically. It is composed of primitive cells, with large, hyperchromatic nuclei, numerous nucleoli, and a poorly differentiated cytoplasm (1).

The growth of this primitive trophoblast into the maternal endometrium is rapid and, to a certain

degree, destructive. Maternal blood vessels are surrounded and invaded. Trophoblastic cells replace the maternal vascular endothelium (2) and penetrate into the maternal blood stream (3). Mitoses are numerous in this actively growing tissue.

A glycoprotein, human chorionic gonadotropin (HCG), also a tumor marker, is normally produced by the trophoblast (4). The aggressive growth pattern, vascular invasion of the host, and the secretion of HCG point toward a certain analogy with malignant tumor growth. Once the implantation of the conceptus is achieved, the trophoblast continues to grow in a less invasive fashion. The trophoblast is separated from the maternal tissues by an acellular layer of fibrin and mucoproteins which has been suggested to arrest any further invasion of the myometrium (5). A differentiation into cytotrophoblast and syncytiotrophoblast further takes place in which the primitive cytotrophoblastic cells produce the more specialized syncytiotrophoblastic cells (6).

The chorionic villi develop from primary villi which are composed exclusively of trophoblastic cells, to secondary villi in which a central core of primitive fetal mesenchyme is formed. Finally, by three weeks of gestation, they develop into tertiary villi in which fetal capillaries also are formed that connect to the fetal circulation. The trophoblast

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covers the chorionic villi (7), and is composed of two layers:

1. *The cytotrophoblast*: Consists of large cuboidal cells that are situated on a basement membrane with defined borders, large nuclei with prominent nucleoli, and features numerous mitoses. The cytoplasm contains large amounts of free ribosomes and relatively few organelles. This cell is obviously equipped for growth and proliferation (7).
2. *The syncytiotrophoblast*: Covers the cytotrophoblast and faces the maternal tissues. It is composed of large multinucleated cells that, at the ultrastructural level, are subdivided into an alpha, beta, and accrual zone.
 - a. *The alpha zone*: Has an apical microvillous border, thus offers a large contact surface to the maternal blood. Numerous pinocytotic vesicles are present on the apical border. They are assumed to represent the site of binding and transfer of maternal immune globulins. A complex channel system, phagolysosomes and various vacuoles represent further cellular organelles which are specialized for maternofetal transfer.
 - b. *The beta zone*: Displays an abundant rough and smooth endoplasmic reticulum, numerous mitochondria, secretory vacuoles and a prominent Golgi apparatus. This zone is specialized in the secretion of steroid and proteic hormones.
 - c. *The basal accrual zone*: Has a more simplified structure resembling the cytotrophoblast to which it is closely located. The cytoplasm contains only few mitochondria and rough endoplasmic reticulum. Polysomes and free ribosomes are abundant. This cellular zone has a proliferative function.

As in other tissues, the trophoblastic cells display an inverse relationship between growth and differentiation, proliferation and specialization. The cytotrophoblast is equipped for the former, the syncytiotrophoblast for the latter (6).

Trophoblastic cells which may be found in the maternal venous system as well as in maternal lungs, mysteriously disappear: they never pass through the maternal pulmonary circulation in normal gestation and do not "settle" in any extrauterine location either (8, 9).

Titers of HCG reach a peak during weeks six to ten of normal gestation. Their values reach those of the titers found in malignancy (4). As pregnancy progresses, these titers decline to relatively low values which persist to the end of gestation (4).

It thus appears that after a tumultuous tumor-

like growth of the trophoblast into the maternal tissue at the beginning of the pregnancy, this growth is well controlled and self-limited both at a local and at a systemic level. The maternal organism develops a defense mechanism against the trophoblastic invasion that is probably closely related to the development of maternal tolerance (nonrejection) of the fetal allograft (5). In normal pregnancy the maternal organism possesses the ability to limit the invasive tendencies of the growing trophoblast and, at the same time, to protect it from rejection. It is still to be determined how trophoblastic antigenic modulation possibly influences maternal immune competence resulting in maternal self defense against trophoblastic invasion. In normal gestation, these mechanisms operate probably at a local and at a systemic level. Local factors at the deciduatrophoblast interface probably prevent lymphocytic infiltrates from rejecting the conceptus, as well as trophoblast from invading the myometrium (5). Systemic factors may prevent the mounting of effective antibodies against paternal antigens, possibly transform them into enhancing ones, and destroy the circulating trophoblastic cells, thus prevent them from settling and growing in distant organs (11, 12).

A delicately balanced interplay between modulating trophoblastic antigens and an adjusted maternal immune competence seems to work efficiently in the majority of pregnancies. Gestation is, of course, no tumor and the maternal organism, although immunologically modified and partially depressed, does not seem to be immunologically suppressed in normal gestation (13-15).

Trophoblastic Diseases

Failures of the still incompletely understood process of modulation and adaptation has been suggested to lead to the development of trophoblastic diseases which encompass a spectrum of pathologic entities ranging from spontaneous abortions to the once most malignant tumor of the female genital tract, choriocarcinoma (Fig. 1). Genetic and immunologic factors are known to be involved in trophoblastic diseases, but their precise mechanism and interrelationship are still poorly understood.

Spontaneous Abortions

These were, in numerous studies, documented to be associated with chromosomal anomalies (16, 17). It was also shown that perhaps immunologic factors may be involved in spontaneous abortions. Thus, blocking factors to paternal antigens which are nor-

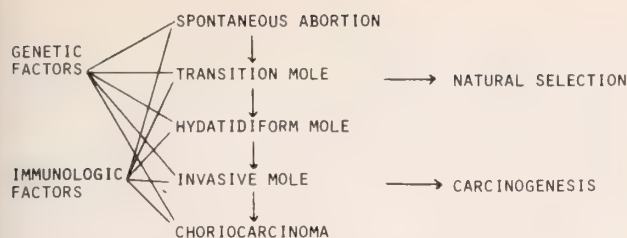


FIG. 1. Schematic representation of the increasing severity of trophoblastic diseases. This does not imply a continuum of disease or progression of a lesion in the individual patient.

mally present in maternal serum, have been found to be absent in habitual abortion (18). The biological significance of the spontaneous abortions as a vehicle of natural selection, aimed at the elimination of chromosomally malformed products of conception, infers thus both genetic and immunologic factors.

Through the study of the morphology of abortion products, it has been suggested that a failure of the trophoblast may be involved in the early sequence of events of the spontaneous abortion (19). It has been demonstrated that the chorionic blood vessels are formed through the angiogenic function of the trophoblast (13). The chorionic blood vessels play a crucial role in the fetoplacental circulation. A failure in their development may interfere with the fetal growth and development or even arrest it. Subsequently, the fetus may be resorbed in the amniotic sac (blighted ovum) (19). If this failure of the chorionic vessels is only partial, and a number of them are functioning, the fetus may stay alive and continue to grow and develop. Chromosomal aberrations are known to be associated with fetal congenital anomalies and phenotypic changes were described in a number of chromosomal anomalies (17, 19). Chromosomal anomalies were also described to be present in trophoblastic neoplasia (20, 21). The morphologic changes of the placenta were in general less satisfactorily defined in the literature than the associated fetal somatic anomalies described as associated with chromosomal aberrations.

In a study performed in our department (Deligdisch L, Strauss L, unpublished data), the histologic features of placenta in cases of documented trisomy, 13 and 18 were investigated. Both conditions were associated with maldevelopment of the trophoblast and the chorionic vessels. The affected chorionic villi showed an edematous loose connective tissue, reminiscent of an earlier stage of gestation, representing an arrest of maturation. Fewer or no chorionic blood vessels at all were found, along with an increase in Hofbauer cells. The latter function is to "Shuttle" between maternal and fetal circulation

and increases in number when the chorionic vessels fail to meet the needs of maternal fetal transfer (22). Another finding in these "dysmature" chorionic villi was the ingrowth of trophoblastic cells from the surface in the inside of the villi probably related to an abnormal angiogenesis (Fig. 2). Affected chorionic villi, alternating with normal ones, gave to the placentas of these chromosomally abnormal fetuses a mosaic-like pattern (Fig. 3).

This study suggested that chromosomal defects are frequently associated with an abnormal development of the trophoblast and the chorionic vasculature, thus probably interfering with fetomaternal exchanges and producing fetal maldevelopment or wastage. It is unknown at the present time if, and if so in what way, the trophoblastic antigenicity is unmasked by the lack of blocking factors normally present in the serum of pregnant women. It is furthermore unknown in which way such an immunologic defect may relate to a following chromosomal defect (or vice versa) which in turn may lead to abortion.

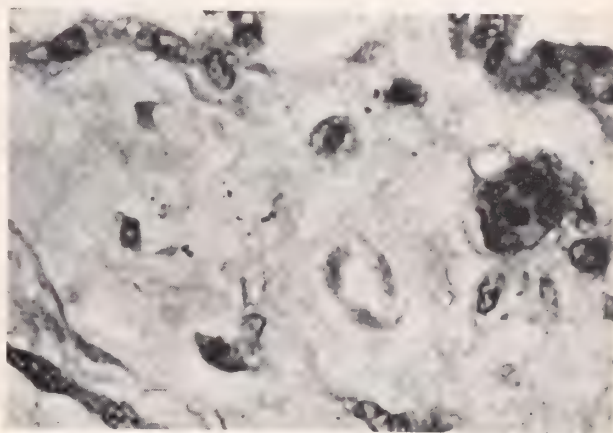


FIG. 2. Inverted trophoblastic cell (right) in chorionic villus with abnormal vascularity, in a case of trisomy 13. H.E. $\times 160$.

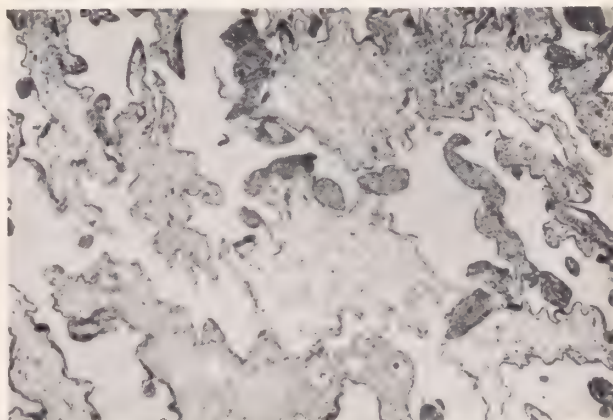


FIG. 3. Mosaic-like pattern of chorionic villi showing normal chorionic villi alternating with edematous, dysmature villi. H.E. $\times 40$.

"Transition Mole"

Another entity within the group of trophoblastic diseases, next in the increasing order of severity, is represented by the "transition mole". The vascularity of the chorionic villi is compromised and the connective tissue undergoes hydropic degeneration in only some portions of the placenta (19). The trophoblastic layers may or may not show hyperplastic changes.

Hydatidiform Mole

This is a disorder well recognized since ancient history. Its geographic distribution in the world ranges between one case of hydatidiform mole per 82 pregnancies in the Far East, to one per 2,500 pregnancies in the USA and Western Europe (23, 24, 25). Although demographic data are incomplete it seems that the etiopathogenetic factors are not racial but are related to socioeconomic and dietary factors, multiparity, and very young or old maternal age (26). All of these factors therefore point toward a weakened maternal host.

The morphologic changes of the hydatidiform mole consist of a grossly very abnormal development of the placenta which is composed of grape-like vesicles and, in the majority of cases, absence of fetus. Histologically, in addition to the lack of chorionic blood vessels and hydropic degeneration of the villi, a marked proliferation of trophoblastic cells is the hallmark of this disorder (27). The hydatidiform moles, in their majority, are diagnosed during the second trimester of pregnancy, when normally the trophoblastic layers consist of a relatively poorly represented cytotrophoblast and a well differentiated syncytiotrophoblast. The trophoblastic layers proliferate in a fashion reminiscent of the early stages of gestation, displaying solid masses of cytotrophoblast and plexiform structures of immature syncytiotrophoblast that are actively growing into the intervillous spaces (25, 27). The increased nucleocytoplasmic ratio, nuclear pleomorphism, dedifferentiation of the syncytiotrophoblastic cells which, during the second trimester of pregnancy normally should become syncytial knots, and the high number of mitoses are reminiscent of both the trophoblastic growth of early implantation and the growth of malignant tumors. Also the production of HCG which normally should be low during the second trimester of gestation, rises to peak levels equal or higher than the normal peak of six to ten weeks of gestation. At this point, it seems that a process is faced in which morphologically and biologically the pathways of degenerative changes in-

duced by genetic and developmental defects are joining those of neoplasia.

The benign outcome of most of the hydatidiform moles has been thought to be due to maternal immune competence and ability to limit the trophoblastic invasion (28). The excellent response to chemotherapy is considered due to a host factor (29-31).

In a recent publication, we described certain morphologic characteristics of gestational trophoblastic neoplasias, including tumor virulence and host response, that correlate with response to chemotherapy (32). By comparing the morphology of trophoblastic tumors that reacted favorably to chemotherapy with those that reacted unfavorably or in which chemotherapy completely failed, we were able to define some characteristics which seemed relevant to the biological behavior of these tumors. It appeared that in the trophoblastic tumors in which chemotherapy failed, the histologic structure revealed a predominantly cytotrophoblastic component which proliferated in solid sheets (Fig. 4). A plexiform pattern associated with the growth of the syncytiotrophoblast, in which syncytial cells form a network of cellular elements with multiple and clumped together nuclei, was found predominantly in those trophoblastic tumors in which one course of single agent chemotherapy had a prompt and favorable effect. We also recognized as an important host factor the presence of the "Striae of Nitabuch," an acellular layer described decades ago at the maternal-trophoblast interface, and more recently proven to be composed mostly of fibrin and glycoproteins (33-35). The presence of this layer at the tumor host interface was correlated with a favorable response to chemotherapy. In the cases of complete failure of chemotherapy, this layer was found to be absent and the trophoblastic tumor cells invaded

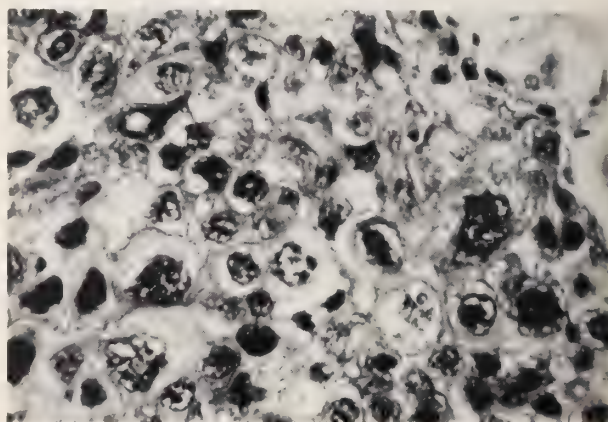


FIG. 4. Solid sheet of mostly cytotrophoblastic cells showing marked nuclear pleomorphism. H.E. $\times 160$.

the myometrium without any separating layer (Fig. 5) (32). The absence of this acellular zone normally separating fetal from maternal tissues and considered to be the site of immunologic masking or enhancement in normal gestation may represent the breakdown of local mechanisms within the maternal defense against the trophoblastic invasion. It seems at this point appropriate to mention that this layer is very much thickened in toxemic pregnancies. In experimental animals, the thickness of the acellular zone increases with the parental dissimilarity (36). The presence of lymphocytes at the interface between trophoblast and maternal tissue was another finding which correlated with a favorable response to chemotherapy. Patients who died of overwhelming trophoblastic tumor and in whom chemotherapy had failed completely, had no lymphocytic infiltrates at the tumor-maternal tissues interface and had also a profound lymphopenia in their peripheral blood (32).

Invasive Mole (Chorioadenoma Destruens)

This form of molar disease is characterized by a deep infiltration of the myometrium by molar chorionic villi with an aggressively growing pattern of trophoblast. It represents a breakdown of local barriers normally present to prevent trophoblastic invasion of the myometrium. Invasive moles may destroy the myometrial wall by perforation and produce a hemoperitoneum. Only rarely does the trophoblastic tissue extend beyond the uterus to other pelvic structures, and distant metastases are uncommon (27).

Choriocarcinoma

This is the most malignant among the trophoblastic neoplasias and was, before the use of chemo-

therapy, a lethal disease. Histologically, it consists of a diffuse invasion of the uterus and other maternal tissues by trophoblastic tissue. Chorionic villi are generally absent. The maternal tissues surrounding the tumor are necrotic and hemorrhagic. They show no lymphocytic reaction, indicating a loss of host defense (37). Patients with choriocarcinoma seem immunologically deprived. Supporting evidence comes from the observation that they have an impaired rejection reaction against their husbands' skin grafts (38).

The microscopic pattern of choriocarcinoma is characterized by an extreme dedifferentiation of the tissue to the most primitive embryonal structure, featuring numerous mitoses and an extreme cellular and nuclear pleomorphism. Choriocarcinoma is usually a widespread neoplasm with involvement of distant organs. Frequently metastases are the presenting symptom (33, 37). By contrast with other widespread neoplasias in which chemotherapy has a limited effectiveness, choriocarcinoma may be, even in the presence of distant metastases, completely cured by chemotherapy. A challenging and incomprehensible feature in the natural history of this neoplasia is the occurrence of spontaneous remissions of metastatic as well as primary tumors (27). In fact, at autopsy, the uterus may not show any tumor tissue in cases of disseminated trophoblastic tumors which led to the patient's death with peritoneal or cerebral hemorrhage (27).

General Aspects

Although histologically frequently identical, it is important to note that gestational choriocarcinoma has a different biological behavior than nongestational, gonadal choriocarcinoma. The generally favorable response to chemotherapy and the occasional spontaneous remissions of gestational choriocarcinoma is not seen with gonadal tumors and may be related to modified host response.

The elevated titers of HCG in trophoblastic tumors reach exceedingly high values in choriocarcinoma. Radioimmunoassays of the beta subunit of HCG represent a sensitive method to detect even small amounts of this hormone (39, 40). As a true tumor marker, HCG in patients with trophoblastic tumors is extremely useful in the monitoring of chemotherapy (41-43). In recent years, an ever increasing number of tumors of various origins have been described to produce HCG. Bronchial, intestinal, ovarian, hepatic tumors, in fact, potentially every anaplastic malignant tumor is capable of secreting HCG.

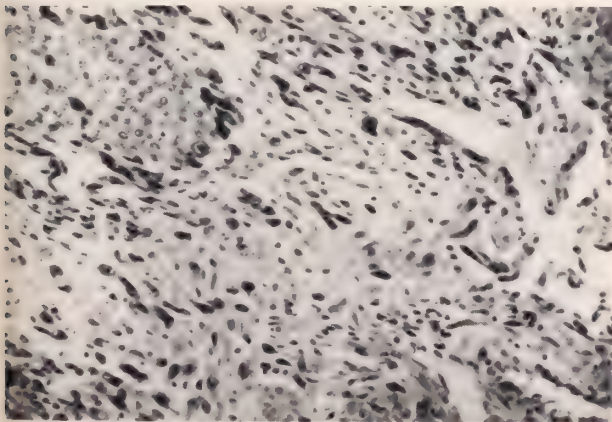


FIG. 5. Myometrium invaded by trophoblastic tumor cells (upper left corner) without separating acellular layer of fibrinoid. H.E. $\times 100$.

Anaplasia in malignancy represents a reversal to primitive cellular characteristics. Its extreme expression is the reversal to an undifferentiated embryonal stage. The production of substances such as CEA and AFP by numerous malignant tumors represent the derepression of substances produced during fetal life by somatic embryonal structures (44, 45). The production of HCG is a further step backwards, to such an early stage of embryonal development that the somatic structures have not been formed yet. The tumor marker HCG is the product of extraembryonal conceptual tissue, the trophoblast. This journey to the past is evidently shorter in choriocarcinoma, a tumor arising in trophoblast which produces HCG in its normal state (4).

Choriocarcinoma is the most severe entity amongst the trophoblastic diseases. Little is known at the present time about its histogenesis and natural history. Thus, it is unknown how the process of implantation and placentation, which usually represents a physiologic self-limited growth of a primitive tissue into an antigenically dissimilar and immunologically modified host, suddenly changes to an aggressive, destructive growth of an anaplastic tissue into an immunologically deprived host. It is unknown what imbalances the physiologic relationship of self-defense against trophoblastic invasion and tolerance of the conceptus by the maternal host, and what transforms the normally self-limited growth of the trophoblast and its differentiation to an organ of transfer and secretion, into the uncontrolled growth and dedifferentiation to the most primitive of all embryonal tissues. Searching for answers to these questions may provide an insight into the challenging analogies between pregnancy and malignancy.

It seems relevant at this point to mention some of the experiments performed by Larsen, who implanted tumor cells from a human choriocarcinoma into hamster liver. He observed at electron micrographs that these very undifferentiated trophoblastic tumor cells bind to the hamster hepatocytes by desmosomal attachments and eventually by fusion, in a sequence of events similar to the trophoblast in early implantation (10).

This observation closes the circle of successive observations relating pregnancy and malignancy.

The study of the spectrum of trophoblastic diseases including elements of trophoblastic virulence and host deficiency, the impact of genetic and developmental disorders on the immunologic fetomaternal interplay, and their relationship to carcinogenesis, may help to gain insight into the natural

history of cancer in general. The trophoblastic tissue is the site where genetical, immunologic and hormonal factors are involved in tumor growth, more obviously perhaps than in any other tissue. Further investigations may benefit both the study of human reproduction and carcinogenesis.

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Immunoassay in Reproductive Medicine

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Abstract

The immunoglobulin is a unique protein that will continue in an ever increasing capacity to serve basic research and medicine. After briefly outlining the development of an immunoassay for human chorionic gonadotropin, this paper will discuss the relatively new discovery in immunochemistry-monoclonal antibody methodology. The importance and application of this methodology are highlighted by a quick review of papers published in the Proceedings of the National Academy of Science during the last fourteen months.

The work on the metabolism of labelled serum proteins by Dr. Solomon A. Berson, former Murray M. Rosenberg Professor of Medicine and Chairman of the Department of Medicine at Mount Sinai, and Dr. Rosalyn Yalow led to the development of radioimmunoassay. This technique is sensitive to concentrations in the picomolar range (10^{-12} M) and specific enough to distinguish between molecules as similar as estradiol and estriol. Indeed, immunoassay in general and radioimmunoassay in particular have been the midwives of much of modern obstetrics and gynecology. Radioimmunoassay has enabled the clinician to obtain reliable measurements of various hormones, proteins, nucleic acids and cofactors; has rendered diagnosis more rapid and more specific; and has opened new approaches to old problems in research. This development in the specificity and sensitivity of analysis of diagnostically relevant biochemicals can be illustrated quite nicely by quickly looking at approaches to the assay of human chorionic gonadotropin over the last 50 years (Table I). With the publication by Aschheim and Zondek in 1928 (1), the era of bioassay of hCG began. This method served as a source of qualitative as well as quantitative information for at least 20 years and is still in use today. In 1944, Jonas Salk published his work on hemagglutination and described this method very carefully (2). In 1951,

Boyden applied a method of protein attachment following tannic acid treatment of sheep red blood cells (3), thus preparing the way for the immunologic assay of hCG published in 1961 by Lief Wide (4). This is the UCG test still in use today. The previous year, 1960, saw the publication of another immunoassay, "Immunoassay of Endogenous Plasma Insulin in Man" (5). The authors were Yalow and Berson. Thus, the era to the diagnostic use of the immunoglobulin was launched. In the late 1960's, RIAs for gonadotropins began to appear with a paper in 1966 on LH by Odell and coworkers (6) and on FSH in 1967 by Midgley (7). Further understanding of the subunit structure of glycopeptide hormones led to the publication of a radioimmunoassay which could distinguish between two similar gonadotropins, LH and hCG. This is the paper by Vaitukaitis and coworkers describing a radioimmunoassay for the beta subunit of hCG (8). Five years later, two groups published papers describing the purification of the tail piece of hCG to which antisera is now available (9, 10). These developments in peptide hormone assay have been paralleled by advances in steroid radioimmunoassay, starting with the work of Liebermann in the late 1950's (11) to the present highly specific antisera to steroid hormones available today.

It is important to recall at this point that radioimmunoassay is basically an antigen-antibody reaction. In fact, the title of the 1960 paper by Yalow and Berson was, as you recall, "Immunoassay of Endogenous Insulin in Man." If the antigen is labelled with a radioactive tag, we call it a *radioimmunoassay*; if the antigen is tagged with an enzyme

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TABLE I
Chronology of Assay of Human Chorionic Gonadotropin

Year	Work	Author(s)
1928	Bioassay of gonadotropin	S. Aschheim, B. Zondek
1944	Simplified antibody titration using hemagglutination	J. Salk
1951	Application of tannic acid methods to sheep erythrocytes	S. Boyden
1961	An immunological method for the assay of human chorionic gonadotropin	L. Wide
1972	A radioimmunoassay for hCG using antisera to beta subunit of hCG	J. Vaitukaitis, G. Braunstein, G. Ross
1977	Isolation and sequence of COOH-terminal fragment of hCG	S. Birken, R. Canfield (Columbia); H. Keutmann, R. Williams (Mass. Gen.)

and/or a fluorescent molecule, we call it *enzyme linked* immunoassay or *fluorescent* immunoassay. As you know, the 1970's produced a new approach to immunoassay which utilizes an enzyme tag rather than a radio-labelled ligand (Fig. 1). An antibody is raised to a given molecule. This molecule is also bound to an enzyme. When the enzyme-linked ligand binds to the antibody, the enzyme activity is inhibited. Still, other methods have been developed using fluorescence or fluorescence-quenching in conjunction with the antigen-antibody reaction. In fact, enzyme-linked immunoassay as well as fluorescent immunoassay are replacing some of the radioimmunoassays simply because they do not involve radioisotopes with all of the ancillary equipment and disposal problems.

However, the exciting news in the field of immunoassay does not arise from the antigen side but

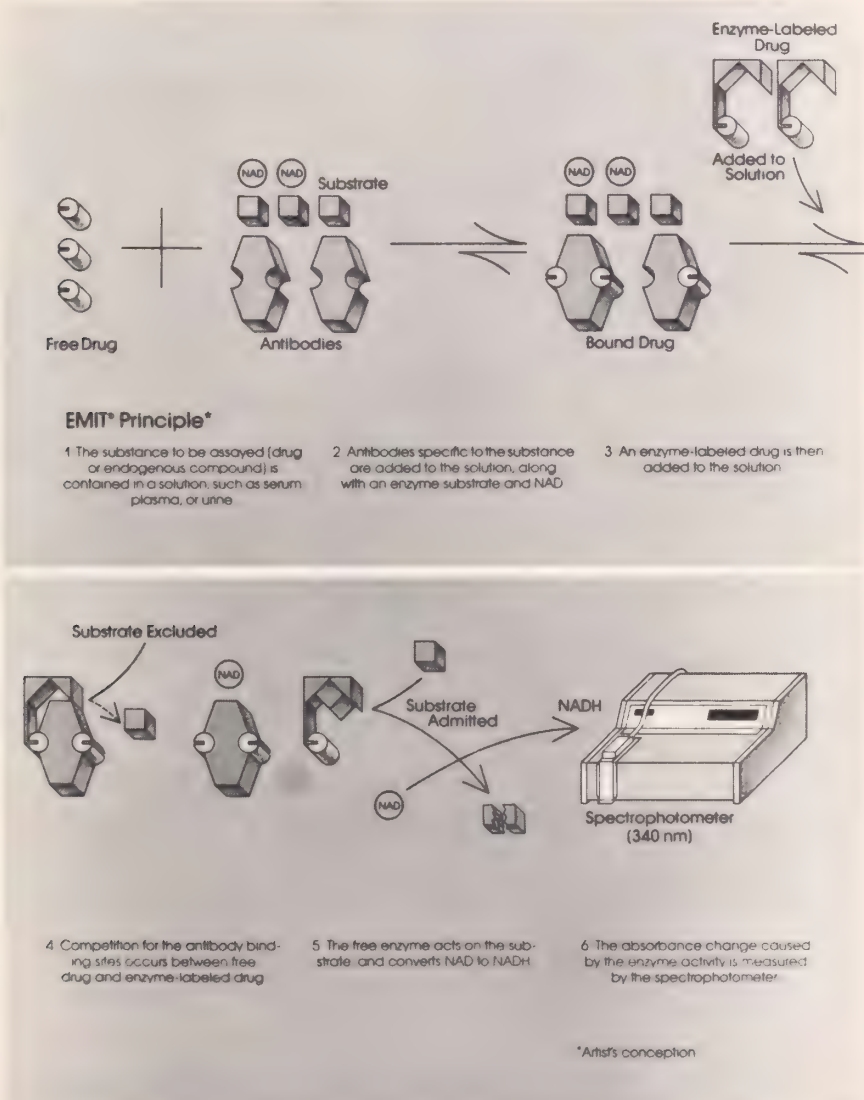


FIG. 1. A schematic diagram of the EMIT[®] principal. NAD and NADH refer respectively to the oxidized and reduced form of nicotinamide adenine dinucleotide. An explanation of each step accompanies the diagram.

rather the antibody side of this fertile antigen-antibody couple.

In 1965, Dr. Leo Sachs and his coworkers at the Salk Institute successfully cultured plasmacytoma cells and produced a line called P_3 , which secreted a large amount of IgG-1 myeloma protein. Although this paper was accepted for publication, it was never published because the cell line was not recovered from the freezer. Horbita and Harris in 1970 re-established a continuous line of P_3 , called P_3K and distributed it to other workers including Cesar Milstein at Cambridge. At Cambridge, a subline of P_3K was developed that had a defective hypoxanthine guanine phosphoribosyl transferase and did not grow on selective medium, that is, medium containing hypoxanthine aminopterin and thymidine (HAT). It was this cell line that produced the first hybridoma from a normal B-cell through hybridi-

zation with a neoplastic cell. Kohler and Milstein published this work in 1975 (12). This method is described briefly as follows (Fig. 2): Antibodies are produced in a mouse in a usual manner. When the titre is acceptable, the animal is sacrificed and a mince of the spleen cells is harvested and mixed with an equal amount of IgG-secreting myeloma cells that have been grown in culture. Fusion takes place in vitro in a few minutes, usually in the presence of additives such as polyethylene glycol. The cell suspension is distributed into multiwell culture trays. Medium containing hypoxanthine aminopterin and thymidine is added. Under these conditions, neither the myeloma cell line nor the spleen cells but only the hybrid cells will grow and form cell clones. Since the cell fusion occurs between spleen cells producing immunoglobulins with specific antibody properties and myeloma cells

SCHEMATIC DIAGRAM OF MONOCLONAL ANTIBODY PRODUCTION

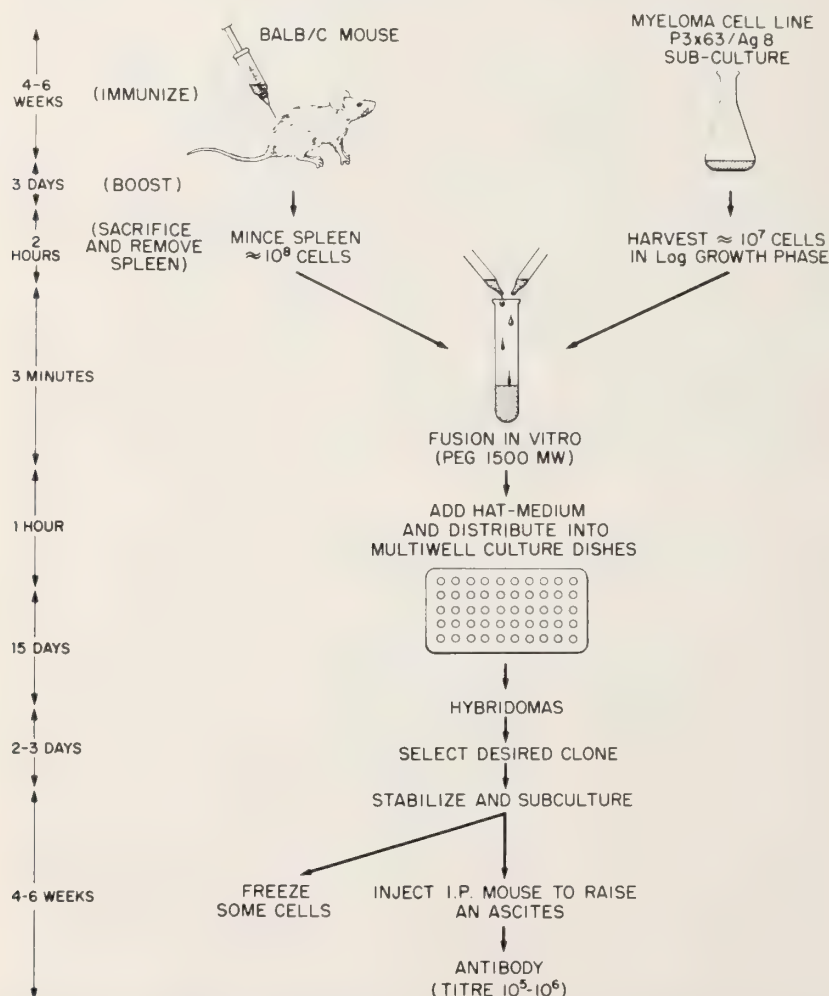


FIG. 2. A schematic diagram illustrating the steps involved in the production of monoclonal antibodies. HAT: hypoxanthine aminopterin and thymidine; PEG: polyethylene glycol.

which secrete IgG in large amounts, the hybridomas that grow are small cell factories secreting predefined antibodies. Within 15 days, one can select those clones which give the antibody titre and specificity desired, stabilize these in soft agar, freeze some of the cells and inject the remainder IP into mice. Because the myeloma cell is a tumor cell, within four to six weeks, one can harvest ascites fluid with antibody titres of 10^5 or 10^6 . Thus in a matter of two to three months, highly specific carefully defined antibody to any protein can be obtained in unlimited amounts. The major importance of this methodology stems from three important factors. First, a monoclonal antibody is a well defined immunoglobulin, not a heterogenous mixture such as is produced from sera of an immunized animal. Secondly, the myeloma spleen cell hybridomas are virtually capable of producing an unlimited supply of this precisely defined antibody. Thirdly, this technique is ideally suited for the preparation of pure antibodies, using nonpurified antigens. This third point needs to be stressed. The exciting aspect of this technology lies in the ability with this methodology to produce antibody probes to individual cell surface antigens (Fig. 3).

It is well known that when an animal is injected with cells from a different strain or species, the antibody response initiated is highly complex and heterogenous. The host will produce an entire spectrum of cell clones, each of which produces antibody to one of the antigens present on the foreign cell surface (13). This is the *raison d'être* of the immune system. It is this transient expression of a single antibody by each antibody producing cell that can be fixed into a permanent hybrid culture through fusion with a myeloma cell. Once fixed, these hybridomas can be purified, stabilized and by fluorescence techniques recently described (14), selected on the basis of their binding to specific antigens. It is this aspect of the Kohler-Milstein discovery which can be applied to the many questions being asked by biologists in general and reproductive biologists and immunologists in particular.

This discovery is to cell biology in general and immunology in particular what the Watson-Crick work in the 1950's was to genetics in general and to biochemistry in particular.

To emphasize its importance, one simply can plot the number of publications in any well refereed journal, for example, the Proceedings of the Na-

SCHEMATIC DIAGRAM SHOWING THE VARIETY OF POSSIBLE IMMUNOGENS

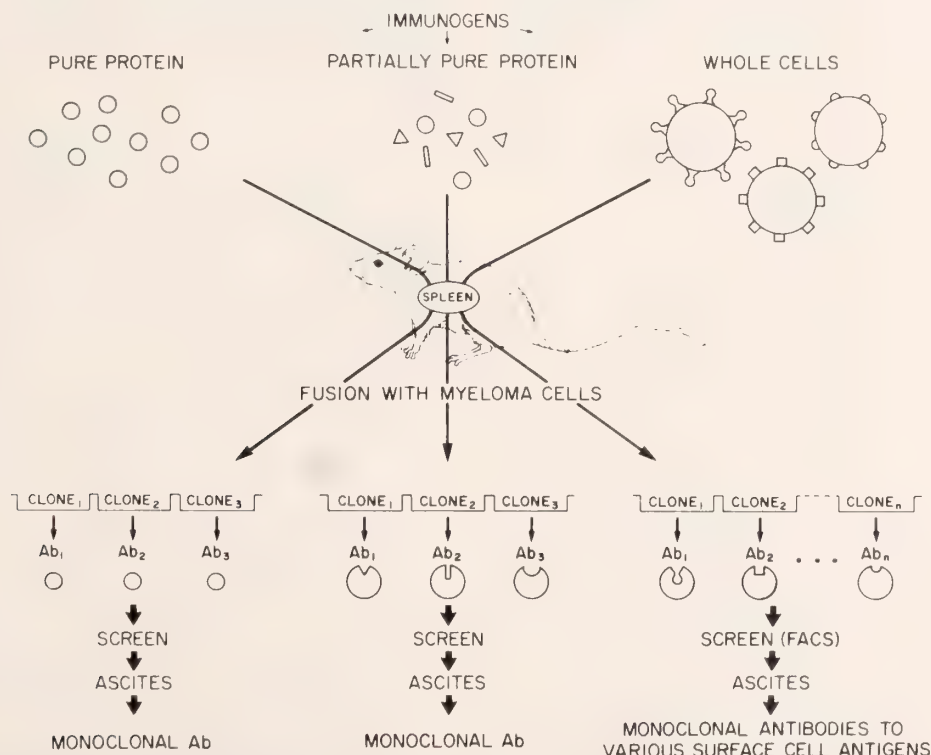


FIG. 3. A schematic diagram illustrating the variety of immunogens possible in the production of monoclonal antibodies. Ab: antibody; FACS: fluorescence-activated cell sorter.

tional Academy of Science (PNAS) over the last four years (Fig. 4). Kohler and Milstein published in August, 1975. In 1976, there were no papers; in 1977, one; in 1978, four; and in 1979, eighteen papers

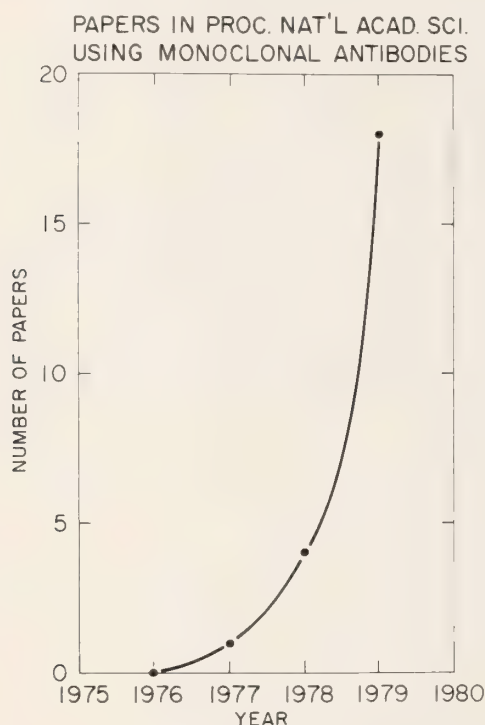


FIG. 4. A graph of the number of papers published in the Proceedings of the National Academy of Science in the last four years in which monoclonal antibodies are described.

were published. To illustrate its application, one simply needs to read the papers published in PNAS from January 1979 to April 1980 (Table II). Elwood Jensen's group has raised a monoclonal antibody to calf uterine estrophilin. This should prove very useful in further understanding of the influence of estrogen on uterine biochemistry. There are three papers dealing with neurobiology, one of them from Cesar Milstein's group at Cambridge, describing a monoclonal antibody to one of the endorphin-like neural peptides, substance P. Antigenic drift in viruses is the subject of the next paper. It is known that such drift results from mutation of viral coat proteins in an immunologically hostile environment. The mechanism is unknown. Understanding of this mechanism can arise from careful peptide mapping of mutants obtained in vitro in a controlled antibody environment. Such an environment can be produced by monoclonal antibodies to viral coat proteins.

Moving closer to the subject of this symposium, two papers have appeared in the last year on the subject of developmental biology. The fascinating phenomenon of differentiation is related to cell-cell interaction. The ability to produce antibodies to the surface proteins involved in cellular communication during embryogenesis could prove to be the key to understanding this phenomenon as well as its aberration, so often seen in metastatic disease. In fact, the disease of cancer and new insights into its

TABLE II

Subject Area	Protein Antigen	Problem Investigated
Endocrinology	Estrophilin	Mechanism of action of estrogen receptor (15)
Neurobiology	Acetylcholine receptor	Subunit structure of this receptor (16)
	Neuronal membrane protein	Neuron-mediated cell-cell interaction (17)
	Substance "P"	Transmission of primary sensory information (18)
		Antigenic drift in type A influenza virus (19)
Virology	Viral surface antigens	Cell-cell interaction in differentiation (20)
Developmental biology	Teratocarcinoma cell surface proteins	
Medical science	Type I collagen	Embryonic development (21)
	Melanoma cell surface antigens	Diagnosis, monitoring, and possible treatment, of melanoma patients (two papers) (22,23)
	Colorectal carcinoma specific antigen	Diagnosis, monitoring, and possible treatment of cancer of the colon (24)
	Human thymus-leukemia antigen	Treatment of leukemia (25)
	Carcinoembryonic antigen (CEA)	Diagnosis, monitoring and possible treatment of adenocarcinoma of the GI tract (26)
Immunology	Hemagglutinin molecule of influenza virus	B-cell clonal response to viral antigens (27)
	MHC gene-linked antigen in rodent human B-cell populations	MHC genetics (28)
	Glu ⁶⁰ Ala ³⁰ Tyr ¹⁰ polypeptide	Mechanism of B cell and T-cell immunity (two papers) (29,30)
	Thymocyte and/or peripheral t-lymphocyte cell surface	The definition and function of human T-cell lymphocytes (three papers) (31-33)
	HL-A/D antigens	The role of the major histocompatibility region and autoimmune disease (34)

etiology can be learned from reading the next four papers in this list. Since certain tumors have already been partially characterized and are available, it is not surprising to see this methodology applied to the study of melanoma, colorectal carcinoma, leukemia and CEA-producing adenocarcinoma of the GI tract. Finally, as we can see from these last five papers, immunology in general and, let us hope, reproductive immunology in particular can benefit from the existence of monoclonal antibody technology.

The December 1979 issue of *Immunologic Reviews* (31) is devoted to a summary of the application of monoclonal antibody probes to an understanding of the HLA or major histocompatibility complex in man. The high resolving power of this technology is contributing to an understanding of these complex polymorphic antigens in the following ways (Table III). First, quantities of HLA antigens large enough for biochemical analysis can be obtained by affinity chromatography, using columns prepared with highly specific monoclonal antibodies. Secondly, identification of *in vitro* translation products of specific messenger RNA can be accomplished by immunoprecipitation from crude RNA preparations using monoclonal antibodies. Thirdly, monoclonal antibodies can be used to study the mechanism of graft rejection by blocking specific surface proteins involved in the immune response. Fourthly, in this vein, monoclonal antibodies could be applied to experiments involving graft enhancement of tissues from closely related nonhuman donors, with the hope of easing the shortage of transplant tissues and organs. Fifthly, xenogenic monoclonal antibodies against the HLA antigens can be used with fluorescent cell sorters to separate fetal cells from maternal blood. In this manner, the crude practice of amniocentesis for karyotyping and genetic analysis during early pregnancy may be unnecessary. Lastly, tumors related to trophoblastic precursors since they lack the HLA antigens, could be detected using fluorescent-tagged monoclonal antibody W6/32 (35).

TABLE III

Application of Monoclonal Antibody Technology to the HL-A Antigen Group

1. Purification (affinity chromatography).
2. Identification of translation products (immunoprecipitation).
3. Mechanism of graft rejection.
4. Graft enhancement for nonhuman tissue.
5. Prenatal karyotyping.
6. Detection and monitoring of trophoblast-related tumors.

In summary, then, the development of immunoassay in general and radioimmunoassay in particular have been presented using the pregnancy-specific hormone, human chorionic gonadotropin as an example. After this brief summary, major emphasis has been placed on the antibody side of the immunoassay couple with a discussion of monoclonal antibody methodology.

Empirical science moves ahead in quantum jumps caused by the impact of methodology. And so, just as the Berson-Yalow discovery has been the midwife of much of current research and medical practice, so the Kohler-Milstein discovery could serve to be the handmaid of future research in immunology and reproductive medicine. When considering the future of reproductive medicine, however, it is worth closing with a quotation from Claude Bernard:

"The spirit of man follows a necessary and logical course in the search for scientific truth. It observes facts, compares them, deduces appropriate results which it controls by experiment, to rise to more and more general propositions and truths. In this advancing labor, a man of science must, of course, know and deal with his predecessors' work. But he must be thoroughly convinced that this work is merely a support from which to go further, and that new scientific truths are not to be found in study of the past, but rather in studies made anew on nature, i.e., in the laboratory" (36).

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Gynecologic Aspects of Malignancies Following Immunosuppressive or Cytotoxic Therapy

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Abstract

Impaired immunity can result in an increased predisposition to malignant tumors. Primary immunodeficiency syndromes, such as ataxia telangiectasia, may result in neoplasia. The incidence of malignancies in organ transplant recipients and in patients treated with chemotherapy is presented. Relative risks of long-term cancer chemotherapy and clinical implications are discussed. The concept of immunologic surveillance in preventing the development of tumors is reviewed.

Introduction

Physicians are becoming increasingly familiar with the use of immunosuppressive and cytotoxic agents. Immunosuppressive compounds are essential for preventing and treating the rejection of organ transplants. The commonly used agents include azathioprine, adrenal corticosteroids, anti-lymphocyte globulin, cyclophosphamide, and actinomycin D. An increased incidence of malignancy has been recognized as a side effect following the use of these treatment modalities (1-9).

In the treatment of ovarian malignancies, gynecologic surgeons use mainly alkylating agents (10). The effectiveness of these drugs in the treatment of ovarian cancer in particular has resulted in a significant number of patients who are at potential risk of developing secondary malignancies.

This paper will review the experience of patients who developed cancer after treatment with immunosuppressive and cytotoxic compounds. We shall consider two groups of patients: 1) renal allograft recipients, and 2) patients with ovarian cancer.

Renal Allograft Recipients With De Novo Malignancies

Human recipients of renal allografts have an increased incidence of de novo cancer development. It was calculated to be five or six percent (or about 100 times) greater than for the general population in the same age range, estimated from 483 cases with a potential follow-up of six months to 11.5 years. The incidence of malignancy development increases in transplant recipients with the passage of time after transplantation (3-5, 7-9). De novo malignancies reported to the Denver transplant tumor registry are shown in Table I. Most striking in this series is a disproportionately high incidence of lymphomas. In the general population, they represent approximately four percent of all malignancies, whereas among transplant recipients, they constitute approximately 24 percent. Possible explanations for the unusual frequency of lymphomas include suppression of certain parts of the reticuloendothelium system by immunosuppressive therapy resulting in overactivity of relatively unaffected parts of this system, perhaps aggravated by chronic antigenic stimulation from the resident allograft. Such overactivity may progress to malignancy or activate latent oncogenic viruses. Others have suggested that the origin of lymphomas may possibly lie in lymphatic cells transferred with donor organs (7, 9, 11). Carcinomas of the colorectum, lung, and

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prostate are observed infrequently in transplant patients (5, 9).

Among gynecologic tumors, cervical intraepithelial neoplasia (CIN) occurs most frequently after treatment with immunosuppressive agents (2, 7). The incidence of CIN is reported as approximately 14 times greater than for the general population. However, invasive carcinoma of the cervix is encountered less frequently (7). In contrast to such cervical lesions, cancers of the endometrium, ovary, and breast are rather uncommon (Table II) (7).

Malignancies in Ovarian Cancer Patients After Immunosuppressive or Cytotoxic Therapy

Twenty-five patients who developed leukemia after immunosuppressive therapy for ovarian cancer have so far been identified in the literature (Table III) (12-19). Eleven of these 25 patients had received radiotherapy in addition to surgery and chemotherapy. Nineteen of the 25 had been treated with single alkylating agents, whereas six had been given combination chemotherapy. The duration of

chemotherapy ranged from ten months to 90 months, depending on interruptions necessitated by drug-induced myelosuppression. The latent periods from onset of chemotherapy to a diagnosis of leukemia ranged from 18 to 114 months.

The true incidence of secondary malignancies in ovarian cancer patients treated with immunosuppressive therapy is unknown. Reimer surveyed 70 institutions using alkylating agents for the frequency of second malignancies. He found an excess of leukemia in ovarian cancer patients treated with alkylating agents, with a relative risk 21 times greater than expected (17). At the Radiumhemmet, 474 patients with ovarian carcinoma were treated with melphalan between 1966 and 1973. Forty-eight patients received at least 300 milligrams melphalan and survived at least three years. Four patients developed acute leukemia (19). Greenspan and Tung reported leukemia developing after chemotherapy in two patients with advanced ovarian carcinoma. These two patients were the only ones out of a total of 400 ovarian cancer patients who had been treated with long-term chemotherapy (15).

TABLE I

*De Novo Gynecologic Malignancies in Allograft Recipients**

Type of Malignancy	No. of Patients
Skin and lip	213
Solid lymphomas	115
Lung	22
Colon and rectum	16
Metastatic-primary site unknown	15
Leukemias	14
Urinary bladder	11
Head and neck	11
Kidney—host	5
allograft	3
Liver and bile ducts	8
Soft tissue sarcomas	6
Thyroid	5
Testis	5
Stomach	4
Brain	3
Prostate	3
Miscellaneous	15
Total	474

* Modified from Penn (8)

TABLE II

*De Novo Gynecologic Malignancies in Allograft Recipients**

Type of Cancer	No. of Patients
Cervix	36
Endometrium	3
Ovary	4
Breast	12
Total	55

* Modified from Penn (8)

Discussion

Immunosuppression has been implicated in the development of malignancies. Loss or impairment of surveillance mechanisms for neoplastic mutant cells have been postulated as a cause of cancer. The high incidence of cancer in patients with congenital immunodeficiency, and in immunosuppressed recipients of renal transplants, has supported this hypothesis. Serious doubts about the validity of immunologic surveillance have arisen because of the following observations: 1) Immunodeficient and renal transplant recipients exhibit a very restricted range of malignancies. Approximately 60 percent of the neoplasms are of lymphoid origin. 2) There is no increased incidence of tumors in several diseases with pronounced immunosuppression, such as leprosy and sarcoidosis. 3) Nude mice do not develop spontaneous tumors. 4) Immunosuppressed mice do not develop spontaneous neoplasms. 5) A variety of cancers do not develop in immunologically privileged sites.

These observations are incompatible with the hypothesis of immunologic surveillance as originally formulated. Thus, it may be that T lymphocytes do not function by eliminating recently appearing tumor cells and, hence, that immunosuppression does not induce spontaneous malignancy through an immunologic surveillance mechanism (20-25).

TABLE III
Reported Cases of Leukemia Following Immunosuppressive Therapy for Ovarian Cancer

Patient	Author	Chemotherapy	Radiationtherapy	Latent period diagnosis-leukemia (Months)
1	Rosner	CTX, TT	9000 R	40
2	Allan	TT	—	30
3	Kaslow	TT, MTX, 5-FU	—	18
4	Greenspan	TT, MTX	—	87
5	"	CTX, TT, MTX	4000 R x 2	114
6	Smit	CLR	—	89
7	"	CLR	—	73
8	Reimer	TT	—	52
9	"	UM, 5-FU	4100 R	26
10	"	CTX	4000 R, P32	30
11	"	PAM	7000 R	39
12	"	PAM, CTX, 5-FU	P32	44
13	"	PAM	2800 R	39
14	"	CLR	5000 R	44
15	"	CTX	—	?
16	"	PAM	7000 R	36
17	"	PAM	?	48
18	"	PAM	?	31
19	"	CLR	—	84
20	"	CLR	—	90
21	Kapadia	TT	4500 R	44
22	Einhorn	PAM	—	33
23	"	PAM	—	58
24	"	PAM	—	76
25	"	PAM	4000 R	35

Abbreviations: CTX = Cyclophosphamide, TT = Thiotepa, MTX = Methotrexate, CLR = Chlorambucil, PAM = Phenylalanine Mustard, 5-FU = 5-Fluorouracil, UM = Uracil Mustard

The exact mechanism of carcinogenesis in man caused by immunosuppressive or cytotoxic chemotherapy is not clear. Chromosomal abnormalities have been described in patients receiving prolonged therapy with alkylating agents (26). Such chromosomal mutation could predispose the patient to develop a second malignancy. Further studies are needed to evaluate the carcinogenic effects that may result from the interaction with radiotherapy and chemotherapy treatments.

The findings described here are important not only for the physician who administers immunosuppressive and cancer chemotherapeutic agents but also for the practitioner who follows the patient's general course. Immunosuppressive therapy should only be used when strongly indicated, as in patients requiring kidney transplantation, and a careful follow-up of patients who are immunosuppressed seems to be essential.

The potential benefits of chemotherapy in the treatment of patients with advanced ovarian cancer clearly outweigh the risk of a second malignancy. In the use of adjuvant chemotherapy, the risks and benefits of chemotherapy must be carefully considered. Patients with ovarian carcinoma who respond to chemotherapy should undergo a second-look op-

eration in order to limit their exposure to unnecessary cytotoxic treatment. Such therapy may be discontinued when the second-look procedure shows no evidence of remaining disease.

For the present, we have to accept the risk of additional malignancies when using immunosuppressive and cytotoxic therapy. Our goal should be to develop therapeutic regimens that are free of potential carcinogenic complications.

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The Immune System in Gynecologic Malignancies

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Abstract

All vertebrates have a defense mechanism, the immune defense system, that protects them from disease-causing microorganisms. Its deliberate exploitation has conquered many infectious diseases and has been a major achievement in medical science in terms of preventing suffering and saving lives.

From the time that Pasteur was able to produce a vaccine against bacterial infections, scientists have attempted to do the same with tumors. Little progress was made until an inbred strain of genetically similar mice was introduced. Up until this time, attempts at immunization resulted in the rejection of implanted tumors, not because of the tumor antigens on the surface of the tumor, but because of the alloantigens that were present. These are much stronger immune stimulators than are the weak tumor antigens.

In 1943, Gross was the first to show that tumor antigens were present and this was confirmed in 1957 by Prehn and Main.

At Lenox Hill Hospital a project was started to study ovarian cancer. This project included ultrastructural examination of the common epithelial ovarian tumors, the tissue structure of epithelial ovarian cancers, and the immunologic study proper, in an attempt to raise a heterologous antibody against a common epithelial ovarian cancer antigen.

The study has progressed to a project in which the isolation of tumor specific antibodies from effusions of ovarian cancer is being explored. Immune complexes have been found in the ascitic fluid as well as on the surface of the cells. These complexes have been eluted by lowering the pH to 3.1 and are separated into two fractions, that is, an antigen and an antibody fraction, by ultrafiltration. Recently, a more effective method using 8 molar urea has been employed. The evaluation of tumor-related activity of antibody preparations by laser nephelometry is underway in the laboratory at Lenox Hill Hospital. This has facilitated the identification of antibody preparations showing significant levels of activity as well as tumor specificity. These antibodies are currently being used as reagents to follow the purification of tumor antigens and to investigate their physical and chemical properties.

All vertebrae have a defense mechanism, the immune defense system, that protects them from disease causing microorganisms. Its deliberate exploitation has conquered many infectious diseases and has been a major achievement in medical science in terms of preventing suffering and saving lives (5).

The progressive and inexorable nature of cancer has lead physicians and laymen alike to believe that humans were incapable of defending themselves against it (47). There was a time, however, when many infectious diseases that are now easily eradicated or cured seemed almost as inevitable in their course as cancer does today. Attempts to fight

cancer immunologically go back to the early days of immunology.

Jenner is considered the father of modern immunology. He observed that milkmaids had smooth skin and complexions whereas the people in the village had pock-marked faces, and, as a result, poor complexions. He suggested that cowpox had given the milkmaids protection against smallpox. He discovered that inoculation with cowpox cross protected man against smallpox during epidemics of this disease. It was fortunate that he chose a related and attenuated virus and did not produce a serious form of the disease, but only a mild disease (6, 11).

Next, Pasteur was able to show that by use of an attenuated bacterial product he was able to protect the inoculated animal against the development of that particular disease. Héricourt and Richet felt that if this could be done with bacteria, the same principle should apply to the control of cancer.

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They injected human cancers into animals and developed an antiserum. When they later used this antiserum, the results were occasionally spectacular but more often, the results were very poor. Some patients developed anaphylactic reaction, others serum sickness and in a few the disease progressed very rapidly. It is obvious to us now, that their serum included cytolytic antibodies that could kill tumor cells and also enhancing antibodies that could cause the growth of tumor cells.

Neither did they realize that humoral mechanisms require antigen, antibody and complement to carry through to the lysis of the tumor cells (8, 23).

In 1908, Paul Ehrlich (15) postulated that aberrant cells were commonly found in the body and that, in the majority of people, these cells remained completely latent thanks to an active immune system. Fifty years later, Thomas stated that the immune system represents a primary mechanism for the natural defense against neoplasia and likened this defense to allograft rejection. Burnet's statement in 1970 that, "immune surveillance is a self fulfilling prophecy," emphasized the etiologic (15) justification for immune surveillance that the "raison d'être" for transplantation immunity is to destroy somatically altered cells that have escaped genetic restraint. The application of immunology to the biology of tumor systems has mushroomed in the past decade resulting in an ever enlarging volume of data relating to the immune responsiveness of patients with malignancy. However, concepts are continually changing and being revised and almost certainly the potentials of tumor immunology are far from being realized.

At the beginning of this century, the hope was held that dissimilarities between normal and neoplastic cells could be demonstrated by immunologic methods making vaccination against cancer a possibility. This concept was based on experiments in which the growth of transplanted animal tumors could be prevented by immunization of recipients of the transplants in various ways. These experiments, however, were performed on tumors originating and tested in non-inbred (outbred) animals. The graft recipients therefore rejected the neoplastic cells not only because of an immunity to (hypothetical) tumor specific antigens, but also—and more importantly—because of sensitization to those normal alloantigens that were present in the animals in which transplanted tumors originated (and, therefore, in the tumor) but not in the recipient. These normal alloantigens are now known as the human leukocyte antigens (HL-A) (20, 21).

When it was recognized that the many claims of tumor specific antigen were based on experiments in which an immunity to normal alloantigens rather than tumor specific antigens had been demonstrated, the field of tumor immunology came into disrepute. A common view was that tumor specific antigens could not possibly exist, since they would confer a selective disadvantage on cells that carry them and lead to their immediate elimination by immunologic mechanism. That neoplasms often metastasize to lymph nodes was viewed as additional indication that an immunologic defense against cancer could hardly play a role.

In 1943, working with so called pedigreed animals that were truly inbred strains of animals, Gross (19) was able to show that mice immunized by intradermal inoculation of tumor cells rejected a subcutaneous transplant of the same tumor, while nonimmunized animals did not. His experiments were done on very small numbers of animals, and there were very few controls so his work was largely ignored. In 1957, Prehn and Main showed that mice immunized against syngeneic methylcholanthrene-induced fibrosarcomas by inoculation of living sarcoma tissue, followed by surgical removal of the growing tumors, were resistant to subsequent grafts of the same tumor. They also showed that immunization with normal tissues did not confer resistance to a tumor graft and that mice that had become resistant to the tumor still accepted skin grafts from syngeneic mates. This work established that there was a tumor specific antigen present on the surface of the tumor cell (17, 40).

It can be said that the modern scientific study of the relationship of immunity to cancer was undertaken in the early 1950's, with extensive work on experimental transplantable tumors and the phenomenon causing the rejection of grafted normal and cancerous tissue in animals. A major conceptual advance in cancer research was achieved when it was established that cancers arouse a specific immune response in organism in which they occur.

Cancer cells, like bacteria and viruses, have their own characteristic antigens. An antigen is defined as a substance—usually a protein or polysaccharide—that the body recognizes as foreign to which presence it reacts by forming antibody. Antigenic differences represent the first known qualitative distinctions between cancer cells and their normal counterparts.

These qualitative differences between normal and cancer cells had escaped other methods of investigation but were revealed by immunologic

techniques that take advantage of the extraordinary power of discrimination of the immune defense mechanism itself. This mechanism is capable of distinguishing even minute differences between protein molecules, probably even one different amino acid in a chain of several thousand (22, 26).

Cancer immunology may now be at the threshold of an era in the prevention and treatment of cancer not unlike that in which the conquest of some major infectious diseases was achieved. Studies in the immunology of cancer have the following objectives: 1) determining the antigenic properties of cancer cells, 2) understanding the details of the mechanism of immune reactions, particularly those evoked by cancer cells, 3) contributing to the knowledge of the etiology of cancer, 4) creating new and better diagnostic methods to increase the effectiveness of all forms of therapy, and 5) developing new therapeutic methods based on an immunologic approach (14).

Etiology

The goal of cancer etiology, the study of the causes of cancer, is cancer prevention. Three types of agents have been shown to cause cancer: chemicals, radiation and viruses. Of these, two—chemical and radiation—clearly cause cancer in man and the third, viruses, are on the basis of persistent knowledge highly suspect. Malignancies may result from a variety of etiologic influences, all of which involve the alteration of genetic code or its expression. These changes are manifested by proliferative advantage, independence of controlling influences, ability to invade, and ability to metastasize. Numerous cell components could be immunogenic under proper conditions, but it is especially the cell surface components that seem most important as antigens. These could be involved as the crucial characteristics of cancer cells and also subject to manipulations that might have influence on the outcome of cancer. A few of the putative tumor antigens described for human tumors to date have been defined in sufficiently, precise and clear serologic terms to be discussed. In Burkitt's lymphoma and nasopharyngeal cancer, tumor specific antigens induced by the Epstein-Barr (EB) virus are present in and on cancer cells. Also, recent studies have shown that tumor specific antigens are present on human malignant melanomas. One class of antigens as specific to the individual melanoma can be recognized by auto-antibody. These antibodies reflect the immune response of the patients against their own tumors (36).

Epidemiology

Cancer epidemiology seeks to correlate differences in incidence of different types of cancer with differences in the external or internal environment of a person or persons developing these cancers. For example, the relationship between cigarette smoking and lung cancer as well as the in utero exposure of the female fetus to diethylstilbestrol (DES).

Silent Cancer

There is evidence that the immune response is an effective defense mechanism that may keep most cancers silent or may even destroy cancer cells in the organism. For example, persons with immunologic deficiency, whether the deficiency is inborn as in agammaglobulinemia or acquired, as by immunosuppression following organ transplantation have an increased incidence of cancer. In animals, all procedures that interfere with the function of specific immunity result in increased susceptibility to both chemical and viral carcinogens. Also in animals treated with carcinogens, the longer the latent period before cancer appears, the weaker its antigens, suggesting that more strongly antigenic cells may have appeared earlier and been eliminated (5).

Some cancer patients can live for long times in apparent equilibrium with their tumors, and a small number of well documented cases of spontaneous regression of advanced cancer exist (31). Both of these phenomenon can probably be attributed to particularly effective immune responses. For example, neuroblastoma, a tumor of young children that originates in the adrenal glands, spontaneously regresses less regularly than other cancers. Recently, immunologic tests on patients in remission have confirmed the existence of an immune reaction in many patients with neuroblastoma. It has also been reported on the basis of studies of the adrenal glands from unselected necropsies on young children, that the incidence of neuroblastoma is 40 to 50 times greater than the overall incidence of clinical neuroblastoma.

Cancers of the kidney, prostate and the thyroid are detected histologically, in tissue samples, more frequently than they are seen clinically. In a series of a thousand necropsies of routine thyroid material, about 50 percent of thyroids showed one or more nodules; of these, 21 percent showed histologic signs suggestive of cancer. Such incidence is very much higher than the death rate for thyroid cancer which is approximately six per million per annum.

Thus, it is apparent that the incidence of silent cancers seems to be much higher than that of overt cancers. Cancer, when first seen by a physician, represents a late stage of a process whose early stages have gone unnoticed. Newly arisen cancer cells are probably commonly eradicated immunologically and, in that event, go undetected. The immune defense system is therefore an efficient surveillance mechanism which, in effect, protects 75 percent of all people from developing the disease. However, there is a challenge to the immunosurveillance concept in cancer. These include: 1) Nude mice and B mice have few tumors without causative infection—they resist chemical carcinogens. 2) Some tumors are prevented by T cell deficiency. 3) Immunosuppression without infection does not necessarily lead to increased cancer. 4) In situ cancer can become large without it initiating immune reaction. 5) Spontaneous and carcinogen-induced cancer may have no demonstrable immunogens. 6) Immunity may be required for development and metastases of some cancer. 7) Chemical immunosuppression—narrow range of cancers. It may be that the immune surveillance mechanism has the ability to eliminate a very small volume of cancer. However, when these tumors because of a tolerance reach a certain burden, the immunosurveillance mechanism is unable to cope with that volume of tumor (50).

Immunologic Means of Cancer Detection

In cancer cells of animals or, more specifically, the surface membrane of these cancer cells, antigens are found associated with DNA and RNA viruses, which are shared by all cancer cells produced by a particular virus. These antigens are referred to as common antigens. Cancer cells produced by chemical carcinogens also have antigens on their cell surfaces, but these are different in each tumor produced by the carcinogen and have been referred to as unique antigens. This rigid separation of antigens has been challenged and now it is believed that both can be present, with one or the other playing the major role (24, 35, 38).

Antigens have also been found in human cancer. In Burkitt's lymphoma and nasopharyngeal cancer tumor specific antigens induced by the EB virus are present in and on cancer cells. A common as well as unique antigen has been found in melanomas. The findings of a common antigen in melanomas suggests a possible viral origin for this type of cancer. An antigen that appears to be a common antigen has also been identified in the common epithelial ovarian cancers.

A new type of antigen, fetal antigen, has been identified in association with human cancers. Fetal antigens are released from the cancer cells and can be detected in the blood stream. These antigens are found in the fetus, most often during the first two trimesters and disappear before birth. They may reappear later in cancer cells. Apparently, during the first two trimesters, a gene is functioning that is repressed in the third trimester or at birth. Later, it is derepressed during carcinogenesis (3, 4, 29).

The two most commonly studied fetal antigens are the carcinoembryonic antigen (CEA) (55) and the alpha-fetoprotein antigen (AFP) (1). Other fetal antigens include leukemia-associated antigen (LAA), fetal sulphoglycoprotein antigen (FSA) and placental alkaline phosphatase. All of these antigens offer promise for the successful development of diagnostic tests.

Of great interest has been the discovery of one particular group of materials, that is, embryonic-specific fetal antigens, in human and animal tumors. It has been noted that these antigens could be released into body fluids. They have been found in spontaneously occurring mouse, rat, and hamster tumors as well as in those induced by physical, viral and chemical agents. It has been proposed that this is a universal oncologic phenomenon termed "retrogenic expression."

Tumor associated macromolecules fall into two groups. The first comprises those that reside within or on tumor cells, which may or may not be released and that may be detected because of their influence on the host's cellular and humoral systems, that is, those present in tumors and body fluids without known metabolic effects, such as, CEA, LAA, FSA, polypeptides and those present in tumors and body fluids and having metabolic effects, such as, placental alkaline phosphatase, placental type hormones, and related products. The second group is constituted of those macromolecules that are not present in or on tumor cells but are released from the tumor cells into the body fluids, that is, the ectopic production of hormones, their subunits and related material. The fetal associated macromolecules are the principal examples.

Cancer Antigens

Just a few of the tumor associated antigens will be reviewed.

Carcinoembryonic antigen (CEA) was first described by Gold and Freedman in 1965. The heterologous (produced in another species) antibodies to CEA were produced by injecting saline extracts of

colon cancer into rabbits. Adenocarcinoma of the human colon did not extend for more than 7 centimeters beyond the tumor. Since the resection always included some of the normal surrounding tissue, it was possible to obtain both normal and tumor tissue from the same person. The tumor then had the same HL-A antigen as the normal surrounding tissue and the one difference was the tumor associated antigen present on the surface of the tumor. Therefore, it was possible subsequently to absorb the rabbit antiserum with the corresponding normal bowel obtained during resection of the primary tumor mass. This permitted researchers to circumvent the problem of distinguishing tumor specific from individual specific antibodies in the final antiserum preparations. Initially, an antigen common to all adenocarcinomas of the bowel was demonstrated by a variety of immunologic techniques. Since then, a similar antigen was found in the embryonic gut and it has been called carcinoembryonic antigen. The antigen was identified in the first two trimesters but was absent at birth. It was suggested that the gene producing the antigen in early embryonic life had been repressed. Later in adult life, when it was found associated with adenocarcinoma of the gut, it was felt that the repressed gene had been derepressed. The stimulus for the derepression may have been a virus, a carcinogen, failure of the immunologic surveillance mechanism, age, or some unknown factor. Carcinoembryonic antigen has been found and associated with a variety of tumors, both endodermal and nonendodermal. Since the serum from patients with both endodermal and nonendodermal derived carcinomas had elevated levels of carcinoembryonic antigen, an antigenic site common to several tumors was hypothesized to exist on carcinoembryonic antigen.

CEA has been characterized as a glycoprotein with a molecular weight of 200,000. Studies raised doubts that CEA is a homogenous material. It appears to have similarities in its activity to blood group A substance. On chemical analysis, it has a repeating unit of *n*-acetyl glucosamine which appears to be partly responsible for the antigenicity of CEA. The correlation between blood groups and malignancies is receiving a great deal of attention at present.

Serum alpha-fetoprotein assays (1) have currently gained an important place in the diagnosis and differential diagnosis of hepatocellular and testicular tumors. Observation that elevated alpha-fetoprotein levels may occur prior to the detection of tumors by other means has important implications. In addition, successful therapy is associated

with the decline in alpha-fetoprotein levels to normal, rising again when the tumor recurs. Alpha-fetoprotein is known to be a product of the human fetal liver, gastrointestinal tract, and yolk sac. It has been shown that the endodermal sinus tumor, which is of vitelline origin, invariably gives a positive test for alpha-fetoprotein. Since the outcome and the disease does not appear to differ according to the level of alpha-fetoprotein, there is a suggestion that a homeostatic control exists for alpha-fetoprotein production in the tumor cells. This does not seem to be true for teratocarcinoma. It has its greatest application in studying hepatomas of the liver and endodermal sinus tumors of the ovaries, as well as teratocarcinomas of the ovary. Besides the alpha-fetoprotein (AFP) there are alpha₂H fetoproteins (Alpha₂HF), beta S fetoprotein, and gamma fetoprotein.

The alkaline phosphatase enzymes (16) are known to occur in serum and are derived from the liver, bones, lung, intestinal tract and placenta. The placenta alkaline phosphatase enzyme does not occur in fetal tissues, blood or serum but does occur in maternal serum during the third trimester of pregnancy. It is never found in the serum of a normal male subject. The enzyme was first detected in a patient named Regan who had lung cancer, and has since been found in association with a variety of cancers. When alkaline phosphatase is present, it is useful to monitor tumor progression or regression and can be detected in malignant serosal exudates. In patients with an elevated serum alkaline phosphatase level, found by routine methods, reexamination should be carried out by fractionation to see if the placental alkaline phosphatase is responsible for the elevation. If it is, then a careful evaluation should be carried out to detect or eliminate a latent neoplasm. This isoenzyme has been found in the serum of 40 percent of patients with cancer of the ovary and 70 percent of malignant fluids from patients with cancer of the ovary.

The chorionic gonadotropin (hCG) and human luteinizing hormone (hLH) are both glycoprotein having similar biologic activity. Both hormones are composed of an alpha and beta subunit. The beta subunit confers immunologic and biologic specificity. The placenta secretes hCG which is normally found only during pregnancy. hLH is secreted by the pituitary and is found in the peripheral blood of both males and females after puberty. Since hCG is normally found only during pregnancy, its detection under other circumstance implies that a hCG secreting tumor is present. Until recently, most antisera to hCG could not discriminate between hCG

and hLH. The development of antiserum specific for the beta subunit has been described and documented. It is specific for hCG. It has been shown that approximately 40 percent of all tumors, including those in men as well as women, secrete hCG with the percentage climbing into the 90 plus percentile among embryonic ovarian tumors and testicular tumors. This finding confirms reports that differentiated somatic cells do carry information for all the potential self phenotypes of the body. This has been demonstrated by the technique of nuclear transplantation into an enucleated egg that can then support the development of a fertile adult.

A purified antigen identified as a ferritin (32) by electrophoretic, chemical and immunologic criteria has been studied by Marcus. It has been found in breast cancers. In the studies carried out in the laboratory at Lenox Hill Hospital, it was found that both the normal ovary and common epithelial ovarian cancer contain ferritin. There was no reaction between the purified ferritin and the antisera raised in the rabbit or the antibody extracted from ascitic fluid of patients with ovarian cancer. It was therefore concluded with additional studies that had been done that the common epithelial ovarian cancer antigen was not ferritin.

Ovarian Cancer Project

An ovarian cancer project was undertaken at Lenox Hill Hospital. It was divided into three main divisions for launching an attack for understanding of the ovary that had developed a malignant potential. The knowledge gained would be directed toward the development of a test for early diagnosis. The study included:

1. Ultrastructural examination of the common epithelial ovarian tumors by the electron microscope.
2. The tissue structure of epithelial ovarian cancers.
3. Immunologic study proper in an attempt to raise a heterologous antibody against a common epithelial ovarian antigen.

Later the project was expanded for the isolation of tumor specific antibodies from effusions of ovarian carcinomas. This will be covered later in the presentation (45, 46).

Ultrastructure of Common Epithelial Ovarian Cancers

The ultrastructure of the various types of ovarian cancers showed heterogeneity within the same tumor with coexistence of more than one cell type

and with variations from differentiation to anaplasia. The coexistence of serous and mucinous cells in ovarian carcinoma, also noted by Gondoz, can be satisfactorily explained by the histogenetic theory of Hertig, which attributes the origin of all epithelial tumors of the ovary to the multipotential ovarian surface epithelium. This was a significant observation in that it suggested that the epithelial ovarian cancers might possess a common antigen. If this proves to be true, and the results of our study indicate that it is so, the possibility of an immunologic test using pooled antigens for the main epithelial tumors moves closer to reality (7, 25).

Tissue Culture of Ovarian Cancer

The culture of common epithelial ovarian cancer cells in vitro has proved to be a most useful method for the comparative study of their morphology and behavior. In addition, tissue cultures are indispensable for any work in tumor virology and immunology. With few exceptions, all tumors showed signs of initial growth in vitro, manifested by cellular attachment to the glass, spreading, and aggregation. Within the next two or three days, more cells attached and spread on the glass, forming small colonies of epithelioid and polyhedral cells. At about ten to 14 days, a clear growth pattern was most often noticeable, becoming better expressed as the cellular density increased. Some of the cultures never grew enough to permit subculturing and died off after two to three weeks, while others grew quite abundantly for a period of several months. It was noted that cultures which exhibited a certain pattern of growth maintained that pattern consistently after many months in vitro, numerous subcultures, and even after freezing and resuscitation. It was concluded from a comparative study of tissue cultures in light and electron microscopy that the ovarian cancer cells have preserved in vitro most of their salient features. In addition, the cells grow in an immune-free environment. These conclusions have been essential in permitting us to confidently use the cultures of ovarian cancers in immunologic studies.

Raising an Antibody to a Tumor-associated Antigen in Common Epithelial Ovarian Cancers

The tumors were brought to the laboratory under sterile conditions within ten minutes of their removal, fixed for light and electron microscopy, processed for tissue culture and either prepared immediately for tissue extracts or frozen for later use.

Tumor extract for immunization was prepared by

using homogenized, pooled common epithelial ovarian cancer tissues in equal volumes of saline at 4°C. The homogenates from the pooled epithelial ovarian cancers were mixed with an equal volume of complete Freund's adjuvant and emulsified by brief sonication at 4°C. Heterologous antisera was raised by injecting the tumor-Freund's adjuvant emulsion subcutaneously in multiple sites once a week for four weeks, followed by a boosting dose at the sixth week and collection of the sera five days later. The resultant antiserum was screened against pooled soluble antigen from normal neoplastic cells in double diffusion studies. Unlike studies with CEA in which normal colon mucus membrane removed 7 or more centimeters from the cancer site can serve as a control possessing the same HL-A antigens as the cancer, this is not possible in studies on ovarian cancer. Therefore, the serum had to be serially absorbed using a variety of tissues and will be discussed later.

The soluble antigen extract of tissue to be used in the immunoprecipitation test and absorption of antiserum were prepared from the previously homogenized tissue, which is sonicated with an equal volume of phosphate-buffered saline pH 7.2, for 60 seconds at 4°C. The resultant sonicates were centrifuged at 48,000 g for two hours at 4°C. The supernatant was stored at -80°C and used as 50 percent tissue extracts, or dialyzed against distilled water and concentrated by lyophilization. Blood substances and other tissue extracts were either prepared for absorption of antisera or were purchased.

The absorption of the heterologous antisera was performed by suspending from 10 mg to 100 mg of lyophilized tissue extract per 1.0 ml of antisera and storing for 10 hours at 4°C. The sera were then centrifuged at 70,000 g for one hour at 4°C. The absorption process was carried out against a variety of tissues. The supernatant was used as the absorbed antisera in the immunodiffusion studies.

The immunodiffusion test was performed by the method of Ouchterlony in either Hyland pattern C plates (Hyland Laboratories, Costa Mesa, California) or Cordis IDF I and II cells (Cordis Laboratories, Miami, Florida). Precipitation patterns were read after one, two and five days.

Results of the Immunodiffusion Test

Figure 1 compares the reactivity in gel immunoprecipitation tests of unabsorbed heterologous antiserum against soluble antigens of ovarian cancer pools on the left and a pool of normal ovaries on

IMMUNODIFFUSION TEST

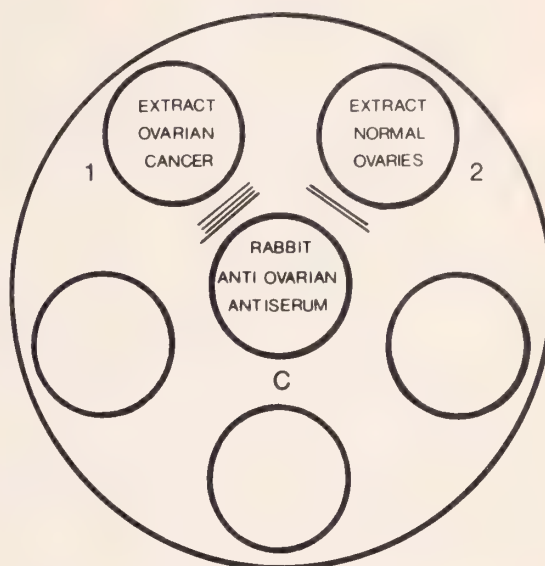


FIG. 1. Immunodiffusion test

IMMUNODIFFUSION TEST

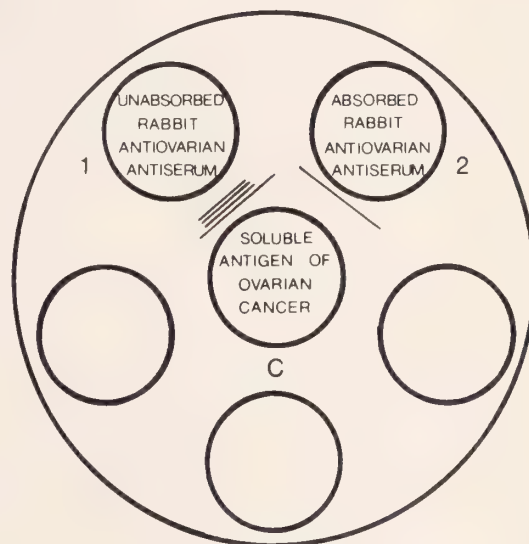


FIG. 2. Immunodiffusion test

the right. An additional precipitation line can be seen against soluble extract of ovarian cancer. The test was repeated using the soluble antigen with both absorbed and unabsorbed sera. The soluble antigen of ovarian cancer pools was placed in the center well and the unabsorbed antisera in well 1 and the absorbed antisera in well 2. Figure 2 is a schematic presentation of the gel immunodiffusion plate. The single line of precipitation could not be

removed by absorption with pooled normal ovary antigens and could only be removed by absorption with common ovarian cancer.

Screening tests using the gel immunodiffusion assays showed that malignant tissue other than ovarian cancer, normal tissue and benign tumors were uniformly nonreactive in the immunodiffusion tests, but all the common epithelial ovarian tests using heterologous absorbed sera gave positive precipitation tests.

Immunofluorescence Test

By using a fluorescent antibody technique, by the indirect method it was possible to show that the antibody identified the candidate antigen in dilution titers that ranged from 1 to 64 to 1 to 1,024. Cells of the common epithelial ovarian cancer maintained in tissue cultures for periods up to one year retained their ability to fix antibody when stained by this procedure.

Isolation of Tumor Specific Antibodies from Effusions of Ovarian Cancer

The observation had been made that if a portion of tumor is removed from the parent tumor and implanted very close to it in the host, the tumor invariably grows whereas, if the tumor is placed elsewhere, in that particular host it very often is rejected. It became obvious that surrounding the tumor was a cloud of substances that offered some protection to the tumor growth. It was therefore decided to explore this possibility.

Peritoneal effusions of patients with ovarian cancer contain sizable amounts of free and complexed immunoglobulins (2). The existence of such autologous tumor specific antibody in ovarian cancer has been shown by work reporting the presence of antibody-antigen complexes (blocking factors) at the surface of human ovarian cancer cells obtained from tumor tissues or malignant effusion fluids. These complexes were eluted at pH 3.1 and separated into two fractions, that is, an antigen and an antibody fraction by ultrafiltration. The autologous anti-ovarian cancer antibodies had potential for use as probes for identification of ovarian cancer antigens in a pure state. The antigen can be used to produce a more specific antiserum in animals. The high affinity antibody raised in animals is essential for developing diagnostic assays such as radioimmunoassays. Although much work needs to be done, the concept of utilization of autologous antibodies as probes for isolating specific ovarian tumor antigens has great potential.

Urea as an Agent for the Dissociation of Immune Complexes

Although a dissociation of antigen-antibody complexes (54) has been previously affected in the laboratory at Lenox Hill Hospital by the use of low pH solution, it is recognized that the method has several inherent disadvantages. The low pH levels, necessary to affect dissociation of complexes (pH 2.8 to 3.0), and a subsequent neutralization represents relatively harsh conditions which may result in significant levels of irreversible denaturation of some IgG molecules, rendering them unfit for use in certain procedures such as complement fixation. However, even such extremes of pH had been found to be ineffective in the separation of antigen from antibody in complexes recovered from the sera of animals bearing experimental tumors. Tucker et al found low pH to be effective at separating anti GP 70-GP 70 immune complexes from the sera of animals bearing a Gross virus induced lymphoma. While low pH is known to dissociate most immune complexes, those with high binding affinities between their antibody and antigen components may be difficult to separate. These and other investigators have turned, with success, to the use of high molarity urea solution (48) for the dissociation of immune complexes and subsequent recovery of active antibody and antigen. Eight molar urea solutions are highly effective chaotropic agents yielding almost complete dissociation of immune complexes. The denaturation of antibodies produced by 8 molar urea is reversible and the removal of urea by dialysis allows the quantitative recovery of antibody and antigen whose physical properties and immunologic reactivities are unaltered. In addition, since urea is a nonionic it does not interfere with the separation of dissociated antibody from antigen by ion exchange chromatography. Chromatography of serum proteins or immune complexes on an ion exchange resin yields pure IgG, as ascertained by agarose gel electrophoresis, with subsequent elution of other protein by high salt solutions. This agent is currently in use in our laboratory at Lenox Hill Hospital and provides a clean and highly reproducible antibody preparation with substantial tumor related reactivity.

At this time antibody preparations have been produced from five ascitic fluids, in ovarian cancer, using 8 molar urea as an agent to dissociate immune complexes. In all cases treatment with 8 molar urea followed by chromatography on QAE Sephadex resulted in immunoelectrophoretically pure IgG with the conversion from high molecular weight

complexes to IgG having a molecular weight concordant with monomeric IgG. The tumor related reactivity of these preparations, in relation to their IgG content is substantially higher than in preparations produced by acid dissociation of immune complexes.

The first of these antibodies has been extensively studied utilizing indirect immunofluorescent tests on sections of various tissues. The antibodies were prepared from a case of poorly differentiated endometrioid carcinoma of the ovary. From 150 ml of ascitic fluid 1.0 ml of antibody having an IgG content of 2.3 mg per ml was produced. The immunofluorescent titer of this preparation against ovarian carcinoma tissue is greater than 1 to 1,000. This preparation was tested against a panel of tissues in a blind study. Using the antibody at a dilution of 1:1000 positive results were obtained with 5 ovarian carcinomas including serous cystadenocarcinomas, endometrioid carcinoma of the ovary, and undifferentiated carcinoma of the ovary. Eight control tissues consisting of carcinoma of the breast, stomach, bladder, lung, colon and malignant melanoma were entirely negative.

In positively stained tumor tissues, only the cytoplasm of tumor cells were stained, the reactive stroma being dramatically negative. This positive reactivity could be blocked by the preincubation of antibody with saline extracts of ovarian carcinoma but not with extracts of lung carcinoma or benign ovarian tumors. In immunofluorescence, the percentage of positively staining tumor cells varies from 30 percent to 100 percent. Blocking experiments using fractions prepared by chromatography of tumor extracts on sephacryl S-200 indicate the blocking antigen has a molecular weight between 70,000 and 100,000 daltons. The high, cross reactive specificity of this antibody preparation, for ovarian carcinoma along with its high titer are extremely promising. Evaluation of additional antibody preparations using a variety of immunologic techniques is currently proceeding in our laboratories.

Evaluation of Tumor Related Activity of Antibody Preparations by Laser Nephelometry

The evaluation of tumor related activity of antibody preparations by laser nephelometry is underway in the laboratory at Lenox Hill Hospital (49). The instrument in use in the laboratory is a Hyland laser nephelometer PDQ. It measures light scattered, from a coherent laser beam by macromolecular aggregates, at 30° forward angle. The sensitivity limit of this system is around 20 ng per ml for a

given antigen. The instrument is equipped with a computer which allows a subtraction of inherent light scatter in both antigen and antibody preparations. In this mode, only the de novo arising light scatter resulting from interacting antigen and antibody is measured.

The reaction is carried out in a polymeric buffer system (4 percent polyethylene glycol 6,000 PBS) at neutral pH. This buffer greatly extends the sensitivity range of the instrument by literally promoting formation of macromolecule aggregates, which gather light, even in conditions of antigen and antibody excess. This system has been applied in the laboratory at Lenox Hill to the study of autologous tumor reactive antibodies and effusions of ovarian carcinoma with very promising results. Antibodies derived from immune complexes in ascitic fluids from ovarian carcinomas show substantial levels of tumor related activity when tested against soluble antigen extracts of tumors. This reactivity is cross reactive between different cases and agrees well with immunofluorescence data. While not all fluids contain recoverable amounts of antibody, those which do, react specifically with extracts of all types of ovarian cancer. All antigen and antibody preparations are pretreated with 4 percent polyethylene glycol 6,000 and ultracentrifuged to minimize inherent light scatter in the polymeric buffer system.

Each new antibody preparation is first tested in nephelometry, over a wide range of dilutions, against a pool of soluble antigen from ovarian carcinomas to determine its level of reactivity. Those showing reaction curves indicative of substantial levels of activity are screened at an appropriate dilution against the panel of soluble antigen extracts from a variety of normal, benign and malignant tissues. In the case of antibodies isolated from effusions of ovarian carcinomas, we have found from 50 to 90 percent higher reactivity against extracts of ovarian carcinomas as compared to corresponding extracts of normal tissues and of malignant tumors of other organs. All extracts are tested at the same total protein concentrations. Reactivity of antibody preparations against controlled tissue extracts can be removed by absorption of antibody without extract in the presence of four percent PEG 6,000. Such absorption results in only small changes in the titer against specific cancer extracts, however, this reactivity can be removed by absorption with extracts of ovarian carcinoma.

Antibody preparations showing significant levels of activity as well as tumor specificity are currently being used as reagents to follow the purification of tumor antigens and to investigate their physical

and chemical properties. The findings in the study of ovarian carcinoma-associated antibodies by laser nephelometry at Lenox Hill Hospital are concordant with previous data obtained by indirect immunofluorescence.

Currently a great deal of work is in progress around the country in attempting to establish the diagnosis of ovarian cancers at an early stage by an immunologic test and/or cure of malignancies by the use of immunotherapy (9, 10, 12, 13, 27, 28, 30, 33, 34, 37).

There has been a great deal of expansion in the application of immunologic knowledge and techniques to the problem of cancer. The field is not static but rather dynamic. Therefore, what is written today is updated by the findings in progress by tomorrow. However, a great deal of promise exists for the serologic diagnosis of early common epithelial ovarian cancer by using an immunologic technique. It is obvious from this review that a variety of immunologic approaches can be applied to the diagnosis of human ovarian cancer and these have the potential advantages of specificity and sensitivity (39, 41-44, 51-53, 56).

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The Immunobiology of the Maternal-fetal Relationship

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Abstract

Many processes of an immunoregulatory nature have been identified or postulated that, operating during pregnancy, could explain the normal exemption from rejection of the fetus as an allograft and its remarkable resistance to specific active or adoptive immunization of the mother. Consistent with all the available evidence is the premise that the success of the fetoplacental allograft depends principally upon two factors—nonintercommunication of its vascular system with that of its mother, and the immunologically privileged status of its trophoblast. Exactly how the latter acquires and maintains its effective nonalloantigenic status has yet to be determined. The significance of other nonspecific factors, such as elevation of various hormone levels, the appearance of certain plasma proteins, such as alpha-feto-protein, known to have immunosuppressive properties, and specific factors such as “blocking alloantibodies” and suppressor cells also remains uncertain.

Maternal/paternal histoincompatibility can be both beneficial and prejudicial to the conceptus: it may augment its chances of implantation and growth rate and it may, through placental porosity to cells, lead to rare instances of maternally acquired graft-versus-host (GVH) disease.

Introduction

Although not an immunologist, because nothing was known about immunology in his time, William Hunter, outstanding anatomist and man-midwife of 18th century London and author of the *Anatomy of the Gravid Uterus* (1774), merits recognition as one of the pioneers of maternal/fetal biology. Through careful dissection and dye injections, Hunter established that the placenta develops from the chorion and decidua and, more importantly, that the circulation of the fetus is entirely separate from that of its mother.

About a century and a half later, with gradual recognition of the consistent fate of tumor, normal tissue, and organ grafts exchanged between unrelated individuals and the development of immunogenetics, came awareness that embryos and fetop-

lacental (F-P) units are allografts with very peculiar properties. In a 1953 classic review entitled “Some Immunological and Endocrinological Problems Raised by the Evolution of Viviparity in Vertebrates,” Medawar (1) succinctly formulated the immunologic problem of pregnancy: “How does the pregnant mother contrive to nourish within itself, for many weeks or months, a fetus that is an antigenically foreign body?”

The initial study that established the transferability of fertilized eggs was also, unwittingly, a classic study on the maternal/fetal relationship. In 1891, Heape (2) procured two fertilized eggs from an Angora rabbit mated to an Angora buck 32 hours beforehand and transferred them to the fallopian tube of a Belgian hare recently mated to a male of her own breed. The subsequent birth of a litter, comprised of four Belgian hares and two Angoras, afforded unequivocal evidence that concepti require no genetic contribution from their mothers to engraft successfully upon the endocrinologically prepared endometrium and survive to term; in transplantation terms, tissue matching appears to be totally unnecessary for the success of the fetal allograft. This conclusion has been reinforced by evidence of the transferability of genetically alien zygotes in many species, and that of tetraparental blastocysts in mice (3).

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An extension of Heape's experiment deserves mention since it highlights the remarkable invulnerability of concepti to an extant state of transplantation immunity. Lanman et al (4) were unable to impair the success of foster pregnancies in rabbits by prior specific hyperimmunization of the surrogate mother against the tissue antigens of both parents of transferred zygotes. Nature's solution for the alien conceptus graft problem is not without limits, the birth of viable offspring following matings between parents of a few closely related species, for example, horses and donkeys, notwithstanding. With most interspecific matings, or inseminations, even if gestation begins, it is usually terminated fairly soon apparently by maternal immunologic reactivity; pregnancies initiated by transfer of zygotes of one species to mothers of another (5) are likewise terminated.

In the 27 years since Medawar's review, the immunobiology of the maternal-fetal relationship has become a very popular and important facet of immunology. The incentives responsible for its development include hopes of: 1) discovery of a new means of immunosuppression applicable to clinical allografts, 2) discovery of an immunologically based method of conception control and of treating certain diseases associated with pregnancy, such as choriocarcinoma, and 3) elucidation of the etiology of certain types of chronic infertility and habitual abortion.

Response of the Gravid Female to Her F-P Allograft

As a baseline for consideration of the mother's response to that special allograft, the F-P unit, review very briefly what happens when a host is confronted with a more conventional allograft, such as an orthotopic skin graft or a fragment of a solid tumor.

Following re-establishment of a blood supply-part of the healing-in process-antigenic material from the graft (or lymphocytes triggered by graft antigens, peripherally) travels via afferent lymphatics to the regional lymph nodes that become the principal seat(s) of the host's response, reflected grossly by lymphadenopathy. A variety of putative effectors is generated. These include alloantibodies specific for graft antigens, such as cytotoxins, that in concert with complement can bind to and destroy target cells and are easily identified by *in vitro* cytotoxicity tests. Other antibodies are produced that are noncytotoxic because of inability to fix complement well. Their role in allograft immunity

is controversial. Some of them bind through their Fc portions to Fc receptors on circulating mononuclear cells which are secondarily endowed with the capacity of inflicting damage upon certain target cells, particularly vascular endothelial cells. This is known as antibody-dependent cell mediated cytotoxicity. Other noncomplement fixing antibodies are probably responsible for the phenomenon of immunologic enhancement, protecting rather than destroying the target graft, that is, enhancing its survival (6, 7).

The cellular mediators generated include 1) cytotoxic T lymphocytes or "killer" cells (Ly1-,2,3+ cells) that can destroy target cells directly by some sort of intimate "kiss of death" that requires neither antibody nor complement, and 2) sensitized T lymphocytes (Ly1+,2,3- cells) which, when activated by antigen, secrete into the local environment a variety of pharmacologically active substances known as "lymphokines." These are potent stimulators of inflammation and thereby recruit nonspecific host cellular defense mechanisms that make a substantial contribution to the rejection process. Thus, the host immune response against an allograft is complex, including both specific and nonspecific components (8, 9). A predominance of activated and killer T cells will procure rejection, as will a predominance of cytotoxic antibodies in the case of certain organ allografts such as kidneys whose vascular supply is surgically restored at the time of operation. However, a predominance of noncomplement fixing antibodies may actually protect the graft, prolonging its life. In the case of primarily revascularized organ allografts, the brunt of the antigenic stimulus probably impinges upon the spleen where the formation of antibodies rather than cellular effectors is more likely to occur (10). Renal allografts are very susceptible to cytotoxic antibodies acting at the level of their vascular endothelium.

Alloantigens

There is considerable quantitative variation in the tissue expression of the alloantigens determined by the major histocompatibility complex (MHC) that function as transplantation antigens. Cells of the lymphoreticular system are particularly well endowed, whereas liver, lung and kidney have moderate amounts and brain and skeletal muscle have very little. Transplantation antigens, which are associated with glycoprotein molecules on cell surfaces, are expressed by both parenchyma and stroma of grafts, including endothelial cells (9). Another important source of transplantation anti-

gens within a graft includes adventitious or "passenger" cells of the lymphoid system and macrophages that populate extravascular spaces and occur as significant minorities in epithelia (11).

Details of the rejection process vary with different types of allografts and with the intensity of the host response, which is determined by the particular histocompatibility difference involved and the host's prior history of exposure to foreign cells. One of the fundamental principles of transplantation biology is that, following exposure of a host to an allograft, it acquires the ability to reject in an accelerated manner subsequent grafts having the same genetic make-up as the original graft.

Consider the impact of a fetoplacental allograft on the mother. As a graft bed, there is nothing very peculiar about the cycling, nonpregnant uterus. It has a good lymphatic drainage, the regional nodes (in rodents) being the iliac and renal nodes. There is, however, some question about the presence of lymphatics in the endometrium (12). Free skin allografts caused to implant of their own accord on the uterine mucosa of nonpregnant female rats are rejected at the same tempo as if they had been transplanted orthotopically, indicating the immunologic competence of the uterus (13). Although a syngeneic pregnancy does not usually prolong the survival of orthotopic skin allografts, it does slightly prolong the life-expectancy of intrauterine skin grafts that may reflect the capacity of decidual tissue to interfere with the afferent pathway of the immunologic response. However, a decidual tissue bed affords an intrauterine skin allograft no respite whatsoever from an extant state of transplantation immunity.

During both syngeneic and allogeneic pregnancies, a pregnant female's lymphomyeloid system displays significant alterations, though the causes have not been well defined (14). Most striking is the transient involution of the thymus in which there is a great reduction in the cortical thymocyte population. Increase in corticosteroid levels, with a possible participation of estrogen, is believed to be responsible. The enlargement of the spleen during pregnancy in some rodents appears to reflect in part significant hematopoietic activity (15).

Medawar's (1) inclusion of "immunological indolence or inertness of the mother" as a possible explanation for the success of the fetus as an allograft has received a great deal of investigative attention. Quantitative changes in the leukocyte population of peripheral blood during pregnancy have been sought with equivocal results so far as alterations in B-, T-, and Null cells are concerned, though

there appears to be an increase in monocytes as well as in phagocytic and bacteriocidal activity (14, 16). Intensive studies have revealed no significant alterations in T cell responses to mitogens or to alloantigens and other antigens in vitro during pregnancy. Tests of general immune function, including reactivity to allografts, have afforded little evidence of significant impairment of a pregnant female's ability to respond by humoral antibody formation or generate a cellular immune response to pathogens or other antigens during pregnancy. The modest depressions of immune responsiveness reported by some investigators have usually occurred late in pregnancy—too late to protect fetal allografts.

Nonspecific Immunoregulatory Substances of Maternal Origin

Many investigators have sought in pregnancy serum or plasma the presence of agents that might diminish lymphocyte function in vivo. Based predominantly upon in vitro tests, this work has identified several classes of candidates that might exert nonspecific controlling influences on T-cells. These include pregnancy zone or PZ proteins, which are alpha-2-globulins, B₁-globulin, early pregnancy factors (EPF), and various hormones, notably corticosteroids, and prostaglandins (17, 18).

Although pregnancy has little effect on the lymph nodes in general, at least in rodents allogeneic pregnancies frequently result in significant hypertrophy of the draining iliac and renal nodes, and the cell population of these organs increases substantially, partly through T cell proliferation and probably partly through "cell-trapping." Only a trivial enlargement of these nodes is usually encountered in syngeneic pregnancies (13). Both in vivo and in vitro studies on cells harvested from nodes stimulated by allogeneic pregnancies in mice have indicated reduction in reactivity to paternal antigens, compared with cells from similar nodes of virgin mice or from nodes stimulated by conventional allografts. A cell-associated suppressor mechanism seems to be involved. In one study, stimulation of the nodes draining the uterus by paravaginal injection of allogeneic tumor cells led to development of cytotoxic T cells, whereas allogeneic pregnancies involving similar alloantigens did not (19).

Apart from the response of a pregnant female's draining lymph nodes to an allogeneic pregnancy, the formation of humoral antibodies to unshared paternal tissue antigens is often evoked. For example, both agglutinating and cytotoxic antibodies appear in the serum of 20%–30% of women during

first pregnancies. The incidence and titres of these antibodies usually increase with the progression of pregnancy and with parity. Mice respond in an essentially similar manner (20).

One of the most striking differences between F-P allografts and conventional allografts is that, despite their ability to evoke certain parameters of allograft reactivity on the part of the mother, F-P allografts are incapable of evoking transplantation immunity, that is, heightened reactivity against test grafts of paternal-type tissue. Indeed, repeated allogeneic pregnancies lead to hyporeactivity on an immunologically specific basis. In part, this seems to reflect the operation of enhancing or blocking antibodies and, in part, the activity of suppressor T cells (21, 22).

Unlike the situation in mice, it appears that in rats MHC-incompatible pregnancies do not evoke the synthesis of hemagglutinating antibodies, though production of hemagglutinins is part of a rat's response to a skin allograft. Beer et al (23) demonstrated that multiparity of Fischer (FI) strain female rats by DA males progressively weakened their capacity to reject skin allografts from (FI $\times$ DA)F₁ donors and produce hemagglutinins on an immunologically specific basis. They found that: 1) excision of the iliac lymph nodes prior to pregnancy enabled the females to produce higher titers of hemagglutinins than intact controls, 2) adoptive transfer of iliac node cells (unlike transfer of spleen or cervical lymph node cells) from parous females to syngeneic virgin hosts was not followed by the appearance of hemagglutinins in their serum, but 3) did lead to a weakened capacity to produce these antibodies when they were subsequently challenged with skin allografts, implicating the iliac nodes as the principal source of the factor(s) responsible for this specific parity-induced, depressed response against skin allografts. That the source of the antigenic stimulus responsible for inducing this hyporeactivity in females was the trophoblast was suggested by the observation that multiple ectopic intrarenal allogeneic pregnancies of pure trophoblast from ectoplacental cones also resulted in hyporeactivity to skin allografts.

Why multiparity leads to hemagglutinin formation in mice, rather than its suppression as in rats, is difficult to explain, especially in light of the evidence that passive enhancement in mice results in suppression of their hemagglutinin response to allografts. Possibly pregnant rats synthesize blocking or enhancing antibody with greater facility than mice, or in the former it is absorbed less avidly by the trophoblast.

In essence, the alloreactivity of a female to a histoincompatible F-P unit differs very significantly from her response to a tissue allograft in that there is a depressed capacity to generate cytotoxic effector cells, a minimal cytotoxic antibody response and an elevated synthesis of antibodies with blocking properties. Activated T cells are generated during pregnancy as are memory cells (14, 18).

Since sensitized maternal cells are present during pregnancy, some factor(s) must prevent either the generation or the expression of their cytotoxicity both in vivo and in vitro. Two agents appear to share this role: 1) enhancing or blocking antibodies that can block cell mediated effector function in several ways, including simple masking of antigens, and 2) suppressor T cells.

Despite the attention that it has attracted, this altered reactivity of females to F-P allografts is not a sufficient explanation for their nonrejection since such females are incapable of harming their fetuses even after active or adoptive immunization with regard to paternal alloantigens, which enables them to reject paternal type allografts in a peremptory manner.

Consideration of the Fetus as an Allograft

The fetus and its associated placenta eventually comprise a relatively large allograft, or perhaps more correctly a parabiont, in the maternal uterus. Direct vascular connections are never established between the two circulations; in their stead there is a very strict quarantining mechanism in which the blood of each partner is dialysed against that of the other. In hemochorial placentas, the dialysis membrane is constituted principally by the syncytiotrophoblast—a relatively enormous area of tissue of fetal origin directly and continuously exposed to maternal blood that may bear both cellular and humoral immunologic effectors directed against surface antigens it has the genetic potentiality to express. The relationship between the syncytiotrophoblast and maternal blood in a fetoplacental allograft is analogous to that between the vascular endothelium of a renal allograft and the host's blood.

Even before implantation, those blastocyst cells destined to give rise to the embryo, per se, express both minor and major histocompatibility antigens on their surfaces (18). Tissues obtained from early embryos and fetuses are both capable of evoking and susceptible to transplantation immunity on the part of their mothers so that there is no question

about the fetuses' need of some form of protection from potential immunologic attack on the part of their mothers.

The Immunologic Properties of the Trophoblast

In 1932, on the basis of inability to detect blood group antigens on human placental villi, Witebsky and Reich (24) suggested that the trophoblast could function as an immunologic protective barrier if its cells were nonantigenic. Subsequent classic transplantation studies on tubal eggs, blastocysts and ectoplacental cones in mice, by Simmons and Russell (25), led to the interesting conclusion that a differentiated trophoblast represents a specialized form of cell that is incapable of synthesizing or effectively expressing on its surface alloantigens displayed by its immediate precursors; that is, it is an immunologically privileged tissue, somewhat like hyaline cartilage.

More recent studies, using a variety of sophisticated techniques, have confirmed the transient appearance and disappearance of both major and minor alloantigens from murine trophoblast during the peri-implantation period (26). Investigations on mature human placentas have virtually failed to reveal HLA-A, B or C antigens, B₂-microglobulins or Ia antigens on the plasma membrane of syncytiotrophoblast (26, 27). By contrast, the mesenchymal cells and vascular endothelial cells within the chorionic villi do express HLA antigens. The once popular belief that a thick sialomucin glycocalyx associated with trophoblast plays an antigen-quarantining role has failed to withstand critical tests (17).

It is important to stress that the trophoblast of the blastocyst, which forms most of the fetal component of the hemochorial placentas of rodents and primates, differentiates into several different kinds of trophoblast cell, not all of which are in contact with maternal blood or solid tissue. Consequently, recent reports of the presence of histocompatibility antigens on dissociated trophoblast cells obtained by disruption of placental tissue do not necessarily mean that such antigens are expressed on the trophoblast cell surfaces actually in contact with maternal tissues (28, 29).

The existence of alloantigens on subpopulations of trophoblast cells could help to account for the high levels of alloantibodies directed against paternal HLA-antigens found in the blood of women suffering from hydatidiform molar pregnancies which are comprised almost entirely of trophoblastic tissue. An alloantigen-expressing component of

trophoblastic tissue may also be responsible for the placenta's ability to prevent the passage of harmful cytotoxic alloantibodies directed against tissues of the fetus, while at the same time facilitating the maternal-to-fetal transmission of beneficial antibodies. It is interesting to note that the placenta does not select against maternal noncytotoxic antipaternal antibodies which may be responsible for the apparent decreased expression of paternal antigens on fetal and cord blood lymphocytes.

The placenta appears to select antibodies for transport nonspecifically via Fc receptors on trophoblast cells and specifically on the basis of alloantigenic determinants associated with certain subpopulations of these cells. Various ingenious maneuvers have been proposed by which some components of the trophoblast might be able to bind and inactivate harmful antibodies, for example, by formation of immune complexes that might be internalized and degraded or shed into the maternal blood stream.

The significance of the trophoblast as a source of the paternal alloantigens responsible for stimulating the maternal immune response still awaits clarification. Immune complexes released into the maternal blood stream might interfere with the central processing of potentially immunogenic fetal alloantigens. Antigen shed directly from the trophoblast, or associated with trophoblastic sprouts in the phenomenon of trophoblastic deportation, may be of significance. It is important to note that exposure to alloantigenic material by the intravenous route favors the production of enhancing rather than cytotoxic antibodies.

Apart from alloantigens, there is a reasonable body of evidence that the placenta possesses organ-specific antigens, associated with the trophoblast (26). Both lymphocytes and antibodies from pregnant and postpartum women display cytotoxic activity against trophoblast cells in vitro without regard to alloantigen specificity (30).

Drs. Beer, Yang and I (31) raised antisera in rabbits against reasonably pure preparations of trophoblast, decidua, fetal, lymphoid and various other tissues from fetal and adult FI rats. After absorption of hemagglutinins, we tested their abortifacient activity in time-mated pregnant rats. Administration of the antisera was via the intramuscular route. The antitrophoblast serum aborted all recipients but had some nonspecific activity since it caused the deaths of 12% of them. The antilymphocyte serum proved to be more toxic for the females though less potent as an abortifacient. An-

tikidney serum caused lesions in the maternal kidneys. None of the other antisera tested harmed either the mothers or their fetuses.

Absorption of the crude antitrophoblast serum with lymphoid cells completely removed its toxicity for the pregnant females without impairing its abortifacient activity. The latter was, however, completely removed by absorption with rat trophoblast cells. This lymphocyte-absorbed antitrophoblast serum proved to be equally effective in aborting rats ranging from 7–18 days postconception; a single dose of 1 ml of antiserum was usually effective. Its capacity to terminate pregnancy was independent of the host's genetic constitution, though its effect was species-specific.

Additional studies on the antigens of the placenta are needed. The information currently available affords some encouragement that control of conception by immunologic means is a feasible objective.

Commencing early in pregnancy, trophoblast synthesizes a number of hormones, most of which have been championed at one time or another as having important immunosuppressive properties (17). Although the serum levels of these hormones are substantial, their concentrations in the vicinity of the trophoblast, in the intervillous spaces, are far greater. Thus, the possibility of their exerting a local rather than a systemic immunoregulatory influence on maternal lymphocytes is worth considering. Progesterone is a particularly attractive candidate for functioning in this manner (32). It is produced by mammalian mothers of all species, irrespective of the degree of anatomical intimacy established in the placenta between the trophoblast and the maternal blood vessels. In high local concentrations this hormone has a powerful anti-inflammatory influence and it can prolong the lives of skin allografts. In vitro, it inhibits mixed lymphocyte reactions and the generation of cytotoxic T cells.

The candidacy of human chorionic gonadotropin (HCG) as an immunosuppressant was compromised by the demonstration that the pure hormone has little or no action on lymphocyte reactivity in vitro. The activity of crude preparations was found to be due to a low molecular weight contaminant. The possibility remains, however, that HCG could function locally by another means to prevent lymphocyte-trophoblastic interaction within the placenta. Present in significant amounts on the surface of syncytiotrophoblast, HCG molecules by virtue of their sialic acid residues may contribute to the maintenance of a high negative charge, repelling

similarly charged lymphocytes. By virtue of their size these molecules might also cause steric hindrance.

The Fetus as a Source of Immunosuppressive Factors

Apart from the theoretical possibility of a direct maternal immunologic attack against the fetal tissue frontier or trophoblast, the fetus as an allograft is potentially susceptible to attack from within by maternal lymphocytes that may have infiltrated across the frontier, that is, graft-versus-host reactivity (33). Recent findings hint at the existence of mechanisms capable of preventing or controlling this hazard. It has been shown that T lymphocytes from perinates can elaborate an agent capable of suppressing the responses of adult lymphocytes (34). In addition, there are various types of suppressor cell in perinates that can alter, by both specific and nonspecific means, various cellular and humoral effector functions of adult lymphocytes (35).

Finally, there is ample evidence of the presence of immunosuppressive substances in amniotic fluid, fetal and neonatal serum that are capable of affecting various immune responses (36). According to some authorities, alpha-feto-protein, (AFP), a glycoprotein produced in the yolk sac and fetal liver, and which is also present in considerable amounts in the maternal plasma, is the principal immunosuppressant agent, or carrier of immunosuppressant agents present in these fluids.

Placental Porosity to Cells—Exchanges Between Mother and Fetus and Their Consequences

The placental barrier is by no means complete or cell-tight. The presence of fetal erythrocytes in the maternal blood is a normal physiologic event, with an incidence of about 30% in the third trimester, increasing considerably thereafter. In instances where Rh(D) negative women are gestating Rh(D) positive fetuses, this cellular exchange may lead to maternal sensitization, the appearance of IgG antibodies in the serum, their transmission across the placenta and the development of erythroblastosis fetalis in the fetus or perinate. The occurrence of this disease has long been upheld as exemplifying the immunologic hazards of being gestated.

There is also evidence that in most human pregnancies leukocytes, including lymphocytes, are transmitted from fetus to mother from as early as the 15th week of gestation (37). However, in hu-

mans it is trophoblastic material, in the form of syncytial sprouts (up to 100,000 per day) that represents the bulk of the fetal donation to the mother. The significance of this trophoblastic deportation which commences as early as the 27th day of gestation still awaits a satisfactory explanation. This traffic in material of fetal origin must be responsible for the maternal response to the alien fetoplacental unit.

Although one might reasonably have anticipated that maternal→fetal cellular traffic would have been negligible because of the hemodynamic relationship between the two circulations, this has not proved to be the case. About 10% of infants have been shown to receive an aggregate equivalent of several milliliters of maternal blood. Except in some cases in which exposure of Rh negative fetuses to Rh positive blood appears to have primed them in utero, exposure to maternal erythrocytes poses no problem for the developing fetus.

Unfortunately, the state of our knowledge concerning the passage of leukocytes from mother to fetus is far from satisfactory in view of GVH disease as a potential hazard of natural infection of fetuses with maternal lymphocytes (33, 38). Experiments on laboratory rodents have shown that carefully timed adoptive or active immunization of females against paternal antigens of their fetuses can lead to the development of high incidences of runt disease among the offspring, presumably due to the passage of effector lymphocytes across the placenta. Indeed, these findings suggest that activated lymphocytes may be better able to cross the placental barrier than unactivated lymphocytes.

It is worth commenting that whenever reactive lymphocytes have been introduced into potentially susceptible fetuses in utero, symptoms of runt disease do not appear until several days postpartum. This observation is consistent with evidence that fetuses have some kind of protective mechanism against this hazard. Overt GVH disease does occur naturally though very infrequently in human infants, and it develops occasionally following intrauterine transfusion of blood to save fetuses afflicted by severe erythroblastosis fetalis.

In animals, mild GVH disease may increase the incidence of lymphomas, and it is of interest that some of the survivors of intrauterine transfusions with persistent chimerism have developed leukemia. A recent observation from studies on experimentally induced runt disease in infant rats is that cerebellar growth is impaired at a time when the animals look perfectly healthy (39).

Histocompatibility Gene Polymorphisms and Maternal-fetal Interaction

From all that has been said so far, nature appears to have evolved a complete and near perfect solution to her allograft problem which applies to a range of placental types; certainly the future of mammals does not appear to be threatened by immunologic forces directed at their concepti. However, 1) the extreme polymorphism of the MHC, 2) the expression of some of its gene products by spermatozoa and most cells of pre- and post-implantation embryos, and 3) specific alteration in the immune reactivity of the mother that occurs in response to these determinants, raise the important question as to the possible significance of histocompatibility genes in mammalian reproduction. The clinical relevance of this question is highlighted by knowledge that spontaneous abortion is the most common form of reproductive failure in humans and that 20% of all abortions are chromosomally normal.

It seems to be reasonably well established that in some but certainly not all human populations, fetomaternal blood group incompatibility can lead to diminished fertility and an increased abortion rate (40). Some cases of chronic abortion could result from a breakdown or failure of immunoregulatory processes that are important for the maintenance of allogeneic pregnancies. One group of investigators has reported that some women with a history of habitual abortions had demonstrable antipaternal cellular immunity on the basis of *in vitro* tests, but lacked IgG antibodies in their serum capable of blocking the expression of that reactivity (18). Such blocking antibodies specific for paternal antigens were usually present in the serum of women who had had normal pregnancies.

Additional evidence of the existence of some relationship between histocompatibility gene disparities and abortion is afforded by anecdotal reports of improved reproductive performance of habitual aborters after changing husbands.

The weak Y-linked histocompatibility locus is interesting with regard to its apparent capacity to interact with other alloantigenic loci. For example, Johanson, Festenstein and Burke (41) found that women who developed HLA antibodies during their first pregnancy gave birth to more male babies than expected, suggesting that male fetuses evoke stronger HLA responses than females. The H-Y system appears to affect the Rh system in a similar way since male fetuses are almost exclusively in-

volved in the exceedingly rare instances of Rh immunization in first pregnancies in women who have had no known abortion or blood transfusion (42).

In experimental animals, two approaches frequently adopted to study the possible influence of histocompatibility genes on reproduction are as follows: 1) to compare the offspring of reciprocal R_2 backcrosses, involving histoincompatible inbred strains of mice or rats, with regard to litter size, sex ratio, survival and segregation ratios of definable histocompatibility antigens, during consecutive pregnancies and sometimes following specific anti-paternal sensitization of the homozygous, parental strain mothers. 2) Using interstrain matings, to study the influence of the maternal immunologic status regarding paternally inherited antigens of the conceptus (sensitized, normal, or immunologically tolerant) on reproductive performance and the fate of the progeny (5, 43).

As are the data from human studies outlined above, the findings reported from these various animal studies are inconsistent and difficult to synthesize. However, with some strain combinations compelling evidence has been forthcoming that maternal-fetal incompatibility and immunologic status with regard to histocompatibility genes can be an important factor in determining the fate of a conceptus (43).

In one of our own studies (13), we found that following local immunization of one horn of a female rat's uterus, by means of a skin allograft, followed by mating with a male against whose tissue antigens the sensitivity was directed, the sensitized uterine horn bore significantly more fetuses than the untreated horn. When a syngeneic mating followed alloimmunization of one uterine horn, there was no disparity, indicating immunogenetic specificity.

In recent experiments in which female FI strain rats were mated to $(FI \times DA)F_1$ hybrid males, we (44) have found that prior splenectomy of the females, followed by sensitization with orthotopic DA skin grafts, resulted in a significant excess of progeny that were heterozygous at the MHC, a decrease in litter size and an increase in the sex ratio.

Milgrom et al (45) have recently reported that with one particular rat strain combination, in which only minor histocompatibility gene differences were involved, hyperimmunization of the females by grafting with paternal strain skin resulted in a significant proportion of stillborn babies. There was also evidence of abnormalities of the epidermis and placental damage. Unfortunately the mediator(s) of these effects was not determined.

Finally, in addition to fetal alloantigens that are capable of immunizing the mother and influencing the fate of her concepti, we must recognize the existence of embryonic or differentiation antigens, which resemble tumor antigens. Since, even in syngeneic pregnancies, these antigens may be foreign to and sensitize the mother, they may be responsible for some fetal wastage. In one study, pretreatment of adult female mice with syngeneic tumor or embryonic tissue, but not normal adult tissue, reduced both the incidence of syngeneic pregnancies and actual litter sizes, and there was evidence that fetal loss occurred early in development (46).

Conclusions

In many disciplines, recognition and interpretation of experiments of nature have played a significant role in their advancement. For example, recognition of the persistent state of cell chimerism that results following early placental fusion and establishment of vascular anastomoses between multiple embryos in cattle afforded the clue that led to discovery of the principle of immunologic tolerance (47).

Choriocarcinoma of gestational origin has been informative for the light it has shed on the unique transplantation properties of trophoblast. Very regrettably, the nonavailability of an animal model of this tumor has restricted the information we can obtain from it (48).

The empirical fact that malignancies are almost never successfully transmitted from mother to fetus may reflect the acquisition of immunocompetence by the fetus long before birth with respect to alien transplantation antigens of its mother, or that the placenta does not readily allow the passage of cells, normal or malignant. A provocative observation that awaits explanation is that malignant melanoma is responsible for almost all (7-8) known cases of fetal involvement by a maternal neoplasm (49).

Since the beginning of this century, many clinicians have entertained the idea that toxemia of pregnancy might have an immunologic component in its etiology, for example, a partial failure of the mechanism(s) responsible for the normal exemption of the fetoplacental unit from attack as an allograft, or a maternal reaction to organ-specific antigens associated with the placenta, especially its trophoblast (50). Although the case that this disease is immunologically based is weak, there is a need for critical largescale studies to determine whether maternal/paternal antigenic disparity, or maternal

reactivity to antigens of the HLA system, are associated with its development.

There is one possible experimental model that merits our serious consideration for the light it may be able to throw on the success of the fetoplacental allograft: It is the long surviving renal allograft in immunologically enhanced rats (Table I) (6, 7). The specifically unresponsive state of enhancement is procurable by pretreatment of the adult host with 1) a relatively small amount of donor alloantigen in the form of a tissue extract or viable cells, either alone or in association with specific alloantibody, or 2) with antibody alone. The antibody must of course be directed against the relevant histoincompatibility antigens of the renal allograft. Established, enhanced renal allografts resemble F-P allografts in that their hosts have: 1) antigraft antibodies and 2) lymphocytes capable of reacting with donor or paternal cells both in vivo and in vitro. Both types of grafts are remarkably refractory to attempts to procure their rejection by active or adoptive immunization of their hosts against donor alloantigens. A considerable amount of evidence suggests that active, ongoing processes in the host are responsible for frustrating the rejection of these two types of allograft. The absence of lymphocytic passenger cells may be a feature common to both types of graft. Although the degree of genetic disparity

TABLE I

Enhancement of Kidney Allografts in Rats

Induced by:

- 1) alloantibody directed against relevant alloantigens of graft (passive enhancement).
- 2) injection of living cells or extracts carrying relevant antigens (active enhancement).
- or 3) combination of above.

Immunologic characteristics of hosts bearing long established renal allografts:

- Partial or complete suppression of specific humoral antibody responses, including cytotoxic IgG.
- Partial suspension of cytotoxic T cell response, unless host re-exposed to donor antigen in form of spleen cells, etc.
- Lymph node cells from enhanced animals will react with donor antigen in GVH assays and in MLC tests.
- Long enhanced renal allografts resistant to active and adoptive immunization of hosts, and to their parabiosis to normal syngeneic animals, but will take up specific antibodies.

Mechanism of passive enhancement involves:

- 1) Inductive phase, in which loss of "passenger cells" from graft may be important, and in which "helper" cells are prevented from being activated.
- 2) Maintenance phase in which MHC antigens in graft progressively induce specific unresponsiveness in absence of activated "helper" cells.

In general, enhancement probably involves modification of the normal interaction between specific lymphocyte subsets, possibly leading to a deficiency in "helper" cells.

TABLE II

Some Mechanisms or Factors Proposed to Explain Exemption from Rejection of Allogeneic Fetoplacental Units

1. *Separation of Maternal and Fetal Blood Circulations*
2. *Afferent Blockade of Immunologic Reflex*
Interference of lymphatic drainage from placenta by decidual tissue.
Avoidance of lymph vessels and nodes by invading trophoblast.
3. *Insusceptible or Neutral Barrier at Maternal/fetal Tissue Interface*
Adaptive failure of trophoblast to express alloantigens effectively, especially where it abuts onto maternal tissue.
Modulation of alloantigen expression by trophoblast in response to maternal antibodies.
4. *Masking of Surface Alloantigens of Trophoblast Cells*
Synthesis of sialomucin coat or thick glycocalyx.
Take-up of agents from maternal blood that result in masking: transferrin that binds to specific receptors, antibodies or antigen/antibody complexes that may bind specifically or nonspecifically (via Fc receptors)
5. *Ability of Trophoblast to Bind Specifically, Internalize and Inactivate Maternal Immunologic Effectors*
6. *Synthesis by syncytiotrophoblast of Agents that Maintain Local Immunosuppressive Environment*
Progesterone, estrogens, HCG, cortisol-binding globulin.
7. *Fetal Synthesis of Immunosuppressive Agents that Enter and Act Within Maternal Circulation*
 α -fetoprotein (AFP); factor released from stimulated fetal lymphocytes.
8. *Maternal Synthesis of Immunoregulatory Agents*
Adrenal corticosteroids
Pregnancy-zone (PZ) proteins, early pregnancy factor (EPF), α -2-globulins
Blocking antibodies
Suppressor cells
9. *Inversion of T and B Cell Numbers in Maternal Blood*

between fetus and mother appears to be of trivial importance in affecting its survival, this is certainly not the case with enhanced renal allografts. The effectiveness of enhancing antibodies declines as the immunogenetic disparity between donor and host increases. Furthermore, when MHC differences are involved, there is a strong gene dosage effect, grafts from homozygous donors being much more refractory to enhancement than grafts from semiallogeneic F₁ hybrid donors.

Heron's (51) demonstration that in rats interstrain pregnancies result in very significant prolongations of survival of auxillary (maternal $\times$ paternal) F₁ hybrid heart allografts is relevant here.

Table II summarizes many ingenious explanations that have been advanced to account for the success of the F-P unit as an allograft. In combination, the complete separation of the maternal and fetal blood circulations and the privileged status of the trophoblast appear to be sufficient in themselves. The remainder represent possible ancillary

or back-up protective mechanisms that may or may not be essential. The exciting challenge remains, however, to determine exactly what principle(s) underlies the effective nonantigenicity of trophoblast.

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Nonneurologic Manifestations of Diabetic Neuropathy

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Abstract

Diabetic neuropathy may involve virtually all systems in the body and, as such, has remarkably diverse clinical manifestations. However, no attention has been directed to tangential, nonneurologic effects of such nerve involvement. These include sterility and infertility secondary to retrograde ejaculation or impotence; chronic urinary tract infection resulting from the incipient asymptomatic diabetic bladder; and vascular events in the brain, heart and feet, conditioned by autonomic nervous system involvement superimposed on existing arteriosclerosis. Orthostatic hypotension may lead to cerebral ischemia and infarction; involvement of the cardiac nerves may lead to myocardial dysfunction and infarction; and peripheral sensory and autonomic nerve involvement play a role in the development of gangrene. As a result of disruption of cardiovascular reflexes, there is interference with homeostatic mechanisms which may precipitate, accelerate, or play a determinant role in vascular events that have become the chief cause of morbidity and mortality in the diabetic.

Interest in diabetic neuropathy has increased rapidly in the past two decades. Many known syndromes of diabetic neuropathy have been more clearly characterized, their pathogeneses delineated and beneficial therapeutic regimes have become available. In addition, new clinical syndromes due to diabetic neuropathy have been described (1). Now, indirect secondary effects are coming to be recognized. These are not caused by neuropathy per se but rather are an accompanying indirectly related consequence or development. This category includes: 1) sterility and infertility, 2) chronic urinary tract infection, 3) vascular events (cerebrum, myocardium, and diabetic foot).

Sterility and Infertility

Retrograde ejaculation occurs in diabetes as a result of diabetic neuropathy involving the pelvic autonomic nervous system (2).

An understanding of retrograde ejaculation depends upon knowledge of the anatomy and physiology of normal emission and ejaculation. Emission

must not be confused with ejaculation; these are two distinct actions. Emission implies the delivering of seminal fluid into the posterior urethra by the sudden contracture of the smooth muscles of the internal genital organs. Under normal circumstances, this is followed by forceful, antegrade propulsion of the fluid from the urethra to the outside by contraction of the bulbocavernosus muscle, and this sequence comprises the normal ejaculatory process (3). Should there be incompetence of the vesical neck, permitting a retrograde flow of propelled material, then one has the phenomenon of retrograde ejaculation. In other words, retrograde ejaculation is a propulsion of seminal fluid from the posterior urethra, retrograde to the bladder through a relaxed internal vesical sphincter. In the diabetic, this phenomenon occurs in the presence of neuropathy involving the pelvic autonomic nervous system, as demonstrated by an abnormal cystometrogram and cystogram and by functional impairment of the internal vesical sphincter. Comparable involvement of the internal vesical sphincter (destruction or inhibition) leads to retrograde ejaculation, which is observed after transurethral resection, after bilateral sympathectomy, as well as in chemical sympathectomy associated with the use of sympatholytic agents (4).

In retrograde ejaculation, orgasm occurs but

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there is no accompanying ejaculate. Following the sensation of ejaculation, catheterization of the bladder yields urine swarming with live, active, motile sperm, thus establishing the diagnosis.

For retrograde ejaculation to occur in a diabetic, there must be several simultaneously operative factors, which include diabetic neuropathy involving the pelvic autonomic system, and maintenance of libido, potency and erectile ability.

Summary: Since retrograde ejaculation results in no sperm reaching the cervix, it is a cause of sterility in the male diabetic (5), and incomplete or partial retrograde ejaculation clearly has an influence on fertility (2). Hence, problems of fertility and sterility in the diabetic require that this possibility be properly evaluated. The induction of pregnancy by obtaining centrifuged postmasturbation urine and injecting it into the uterus has been demonstrated and is encouraging (6).

Impotence

Impotence is impressively prevalent in diabetic men, approximating 50%–60%, which is considerably greater than in the nondiabetic population (7); this increased frequency has been long recognized (8).

Penile erection takes place as a reflex. Stimulation of the second, third and fourth sacral components of the parasympathetic nerves (the *nervi erigentes*), which may be psychic or reflex, results in dilatation of penile arteries, increase in blood flow and tumescence of the corpora cavernosum and spongiosum. Erection is thus induced by active hyperemia of the arteries. Detumescence results from vasoconstriction of the penile arteries with subsequent decrease of arterial blood flow, diminution of pressure and release of compressed veins.

Obviously then, a significant factor in potency is determined by the integrity of autonomic nervous system which is so frequently involved in diabetic neuropathy. Studies have shown a marked correlation between the concurrent presence of diabetic impotence and the involvement of the *nervi erigentes* by diabetic autonomic neuropathy (9).

It is essential to recognize that impotence in the diabetic can and does occur from all causes that affect nondiabetics, in addition to the neurologic involvement of diabetes. Over 90% of nondiabetic impotence results from psychogenic factors and this, of course, may play a role in the diabetic. The differential diagnosis is usually not difficult. Psychologic impotence is typified by its selectivity and periodicity. Additional differentiating factors in-

clude the presence of morning erection, the ability to masturbate successfully, and the occurrence of nocturnal emission. Psychologic impotence usually occurs suddenly, but in diabetes it is a slow ongoing process. In those cases in which the picture is not clear, the use of a mercury strain gauge for measuring the circumference of the penis and monitoring nocturnal erection has helped in the differential diagnosis.

Endocrine factors have long been considered to be the chief cause. However, this has been excluded by the demonstration of normal amounts of plasma testosterone in diabetics with impotence. Furthermore, extensive and intensive therapy with testosterone in its various forms is of no avail.

The clinical picture, as predictable from the pathogenesis, is the striking persistence of libido despite the absence of potency. Another significant clinical feature is that impotence may be the first clinical manifestation of diabetes. Consequently, when a middle-aged man who has previously been potent develops impotence, one must suspect the possible presence of diabetes and take the necessary diagnostic steps.

All patients with diabetic impotence benefited from the knowledge that this manifestation was organic and hence not a reflection of their virility and manhood—a source of great insecurity. Impotence is often the basis of intense marital friction and distrust which may lead to separation and even divorce.

When impotence results from poorly controlled diabetes, it is a reflection of the associated malnutrition and weakness. In such cases, proper management of the metabolic aspects of the disease which restores normal health and vigor also restores potency. If the impotence is psychogenic in origin, the approach must be based on psychologic evaluation and psychotherapy. If drugs are involved, their elimination is usually followed by return of potency. If there is an endocrine basis, the appropriate hormonal therapy should be instituted.

In most cases the cause is neurogenic. Unfortunately, there is as yet no prophylactic or therapeutic modality for neurogenic impotence; hence, the prognosis in these cases is poor. Mechanical devices, including a silicone prosthesis, may be implanted surgically (10). Investigations are now being conducted on an implantable silicone rubber prosthesis, externally controlled by means of a subcutaneous pump located in the scrotum (11).

Since impotence is a cause of sterility, its frequency in diabetic males is significant.

Chronic Urinary Tract Infection

Neurogenic vesical dysfunction with paralysis has been delineated as a neuropathic manifestation of diabetes. It is associated with an atonic distended bladder with residual urine and is inevitably accompanied by secondary infection leading to ascending pyelonephritis, sepsis and azotemia (12). In these instances, chronic urinary infection and chronic pyelonephritis are readily understandable. However, this is the end result of a progressive developmental process beginning with incipient asymptomatic stages. Investigations have shown that there is a frequent occurrence of a detectable incipient asymptomatic stage of bladder involvement in diabetes which presumably leads to the development of the known diabetic paralytic bladder (13). The sequence of events is as follows: a diabetic patient with neuropathy will develop an objectively demonstrable bladder involvement in about 83% of the cases. This is demonstrated by an abnormal cystometrogram and increased bladder capacity. At the onset, there is no residual urine, no growth on culture of the bladder urine, the blood urea nitrogen is not elevated, pyuria and bacteriuria are absent, and albuminuria is either absent or present only in modest amounts. Eventually the bladder decompensates and pyuria, bacteriuria and gross albuminuria are uniformly present.

The fundamental differentiating factor between the incipient stage of neurogenic bladder and the advanced stage of the classic neurogenic bladder paralysis is residual urine. When the incipient stage decompensates, the presence of the abnormal residual urine and stasis lead directly to secondary infection manifested by pyuria, bacteriuria and pyelonephritis. The mere presence of the incipient asymptomatic stage of the diabetic bladder is an inadequate explanation for an increase in pyelonephritis; it is only as the condition advances and urinary retention takes place that there is superimposed infection and ascending pyelonephritis.

Of great theoretical importance is the suggestion inherent in these findings that this is probably the explanation for the increased frequency of urinary tract infections, including pyelonephritis, in the diabetic patient.

It is an accepted fact that pyelonephritis is more frequent in the diabetic as compared to the nondiabetic. This has been confirmed in necropsy material (14), in clinical findings (15), in asymptomatic urinary infection of the diabetic (16), and by determining the bacterial colony counts obtained on

urine culture (17). One of the proposed explanations for this increase is that there is a decreased resistance in infection in diabetes. This is contrary to modern clinical experience. The diabetic has a normal resistance to infection provided that he is under a reasonable degree of control and not in ketosis.

Summary: The increased incidence of urinary tract infections in the diabetic is a result of neuropathy involving the genitourinary tract.

Vascular Events

The dramatic reduction in deaths from acute metabolic derangements that followed the introduction of insulin in 1921 was accompanied by an equally dramatic increase in the longevity of the diabetic patient. Unfortunately, this has been associated with an increasing risk for the development of vascular complications. At the present time, the predominant causes of morbidity in the diabetic patient are lesions associated with macroangiopathy, that is, arteriosclerosis involving the brain, the heart, and the feet.

Arteriosclerosis in diabetes is characterized by increased frequency, earlier onset, accelerated progression, more extensive involvement, a lack of correlation with the severity or the control of the carbohydrate metabolic abnormality and obliteration of the sex difference in that the diabetic female is at equal risk with the diabetic male in this regard. The remarkable disparity and frequency of these complications in the diabetic versus the nondiabetic person has yet to be satisfactorily explained, especially in the absence of any demonstrable histopathologic differences in the arteriosclerosis.

There has been little progress in the ability to prevent, minimize or decelerate these vascular complications. Accumulated knowledge relative to carbohydrate and glycoprotein metabolism has not yielded measurable beneficial effect. Arteriosclerosis, however, is not the only link between diabetes and its cardiovascular complications. There is an increasing awareness of the occurrence of autonomic nervous system neuropathy in the diabetic and its deleterious effects on physiologic functions. This could play a role in pathologic events by interfering with neurogenic and hormonal factors and adversely affect cardiovascular hemodynamics.

The responses to environmental needs in the brain, the heart and the foot are mediated via reflexes that are controlled through the autonomic nervous system. Any change in demand for blood

supply will be reflected promptly by reflex action of the autonomic nervous system. However, if the autonomic nervous system is impaired, and the reflexes are either impeded or disrupted, the normal mechanisms for response to such demands become inoperative.

The Cerebrum

The blood supply to the brain is most carefully maintained. When blood pressure falls, for whatever reason, several mechanisms come into play insuring an adequate cerebral blood flow. Reflex venous and arteriolar constrictions are necessary for the maintenance of the cardiac output and arteriolar pressure in the upright position (18). These reactions are mediated through the baroreceptors which are located in the arch of the aorta and the carotid sinuses (19). They are stimulated by the fall in blood pressure or with upright posture, sending signals via the vasomotor center of the brain to the autonomic nervous system resulting in increased cardiac output and vasoconstriction and restoring blood pressure to normal levels. With more severe decrease in the blood pressure, as occurs during shock, for example, chemoreceptors in the carotid and aortic bodies respond to hypoxia and hypercapnea producing vasomotor constriction via the autonomic nervous system resulting in an increase in blood pressure (20). Thus, for example in health, blood and urinary catecholamines, namely adrenalin and noradrenalin, increase with the change from horizontal to the vertical position. In addition, sympathetically mediated renin becomes available from the kidney.

Autonomic neuropathy in diabetes interferes with the transmission of reflexes and may significantly interfere with these mechanisms. Involvement of sensory baroreceptors, reduction of catecholamines, the lack of response to massage of the carotid sinus to evoke change in pulse rate or blood pressure, abnormal handgrip response, lowering of plasma renin, failure of plasma renin to increase and the absence of compensatory tachycardia have all been well documented. Failure of these mechanisms leads to orthostatic hypotension. Thus, the absence of reflex venous and arteriolar constriction is responsible for the marked impairment of postural adjustment leading to syncope. The absence of compensatory tachycardia in patients with true orthostatic hypotension is one of the major factors leading to vertigo and syncope secondary to a decrease in blood flow (21). Other evidence indicates that pressor catecholamine insufficiency correlates

with postural hypotension in familial dysautonomia (22). In patients with postural hypotension there is little or no increase in blood and urinary concentration of catecholamines—a clear indicator of failure in the chain of events leading to necessary vasoconstriction (23). The change under these conditions from the horizontal to the vertical position is not accompanied by an increase in blood or urinary catecholamines. Diabetic individuals with orthostatic hypotension have a significantly lowered plasma renin-like activity (24).

Summary: Interference with the normal regulatory homeostatic mechanisms by autonomic neuropathy is the basis for orthostatic hypotension leading to diminution of cerebral blood flow resulting in cerebral anemia and syncope. In severe instances this could well lead to transient ischemic attacks and even cerebral infarction.

The Heart

The differences between coronary arteriosclerotic heart disease in the diabetic as compared with the nondiabetic are an increased incidence in both dead and living patients; a striking increase in coronary heart disease in the diabetic female including those who are normotensive, premenopausal, and free of significant hyperlipidemia; a higher early mortality rate; a shortened life among diabetic survivors of acute myocardial infarction; and a higher incidence of absence of pain, that is, asymptomatic coronary thrombosis with acute myocardial infarction in the diabetic individual. Effects of autonomic nervous system neuropathy on cardiac function in the diabetic patient are numerous and well-documented. These are measured by relatively simple methods that are noninvasive, nonpainful, and may at times even be done at the bedside. They include the demonstration of an abnormal Valsalva maneuver (25), absence of compensatory tachycardia (26), resting tachycardia (27), loss of beat-to-beat heart rate variation (28), the lack of response of the pulse rate or blood pressure to massage of the carotid sinuses (29), a failure of the heartbeat to accelerate after intravenous atropine (30), abnormal handgrip response (31), arrhythmia (32), cardiac arrest (33), and histopathologic evidence of autonomic cardiac nerve involvement both on light and electron microscopy (34).

One of the most characteristic indications of autonomic involvement is the loss of beat-to-beat variation of the heart rate during both quiet and deep breathings; this has been demonstrated on several occasions. Most studies agree that the R-R

interval variation is a better indication of autonomic nerve damage than the R-R interval itself (35). Also, resting tachycardia has been documented; in one report, three out of six cases had a heart rate of 100 to 110 beats per minute at rest (36).

The resting tachycardia, the failure of the heart to accelerate normally after intravenous atropine, and the absence of reflex bradycardia as well as the absence of sinus arrhythmia and the decreased variation of the R-R interval all clearly point to involvement of the vagus nerve. As a consequence, there is an added tendency to cardiac dysrhythmia including spontaneous cardiac arrest. One patient with a totally denervated heart had tachycardia, a fixed heart rate, and a tendency to cardiac dysrhythmia (37). In one study spontaneous arrest in three patients with severe autonomic neuropathy was observed (38). It is a well-recognized, clinical observation that has been studied objectively that there is an increased occurrence of either painless acute myocardial infarction in the diabetic patient or a significant blunting of pain. In one study of 100 diabetics with acute myocardial infarction, 42 were asymptomatic; of 100 nondiabetics, only six were asymptomatic (39). Involvement of the autonomic nervous system as the afferent part of the reflex arc has been established by the finding of advanced pathology of the autonomic nerves at autopsy in diabetic patients with painless myocardial infarction (40).

Summary: Resting tachycardia suggests increased demands on the heart resulting from the extra persistent pumping effort required. All the autonomic neuropathic effects help to increase the inability to alter the rate and amount of blood flow from the heart and response to the physiologic and nutritional demands of the tissues. The possibility that this imbalance may contribute to the increased morbidity and mortality rates of myocardial infarction in the diabetic is definitely raised as is the possibility of increased association of abnormal rhythms.

Diabetic Foot

The diabetic foot is characterized by wet gangrene indicating the presence of at least some available blood supply as distinguished from the mummified gangrene of the nondiabetic individual which results from virtually complete absence of peripheral blood supply. In addition, about 10% of amputations performed in the diabetic take place in the presence of palpable pulses. These facts denote that in the diabetic, factors other than peripheral vas-

cular insufficiency alone are involved in the events that lead to amputation. On further analysis, it is evident that the one outstanding difference between the diabetic and the nondiabetic foot is the presence of neuropathy in the former and its absence in the latter. It is well-recognized that the sensory impairment of diabetic neuropathy permits unperceived minor trauma to occur which can lead to secondary infection, gangrene, and amputation (41). This is enhanced by fluid loss, perspiration, dryness and cracking of the skin. Also, there are important abnormalities of vasomotor function which have been heretofore inadequately appreciated in the development of these complications and loss of vasomotor response in diabetic persons with neuropathy. For example, in patients with peripheral neuropathy, there is no increase in blood flow after heating. However, an injection of tolazoline which acts directly on the smooth muscle of the vessel results in a sharp increase in the blood flow, indicating that this defect is neuropathic and not due to occlusive vascular disease. Changes in the skin temperature and by the starch-iodine test have been demonstrated with autonomic neuropathy (42). Blood flow patterns that indicate the presence of varying undulating beat-to-beat change of arterial flow have been measured. However, in diabetic patients with peripheral neuropathy there is complete loss of such spontaneous variations indicating that in the presence of peripheral autonomic neuropathy the ability to make necessary physiologic accommodations to maintain homeostasis, and to meet local demands of tissues that vary from moment to moment has been severely impaired or lost (43).

Discussion

The presence of symptoms in diabetics that are attributable to autonomic nerve involvement have been documented for many years (44). These include sweating disturbances, abnormal temperature changes in the extremities, postural hypotension, vasomotor and trophic signs, and early vasomotor disturbances.

In recent years important advances have been made indicating the impact of autonomic nervous system involvement in regulatory phenomena of the cardiovascular system. The availability of sophisticated tests to assess functional abnormalities that are noninvasive, nonpainful, relatively simple in performance and remarkably informative have been highly instrumental in establishing a significant body of information.

Anatomic evidence of autonomic nerve involvement is available. Employing ultrastructural quantitation of the terminal innervations of small caliber blood vessels in patients with long-standing diabetes mellitus, a group of investigators observed that the nerve findings are either entirely absent or are found at significantly greater distances from the affected sites than in controls (45). They infer that the findings account for some of the vasomotor disturbances seen in diabetic patients such as those with postural hypotension. Anatomic evidence of a lack of end organ innervation in the peripheral blood vessels may be an indication that autonomic sympathectomy occurs in diabetes mellitus.

Implications: The above facts indicate impairment of the ability to respond in normal physiologic fashion to demands placed on the cardiovascular system in the diabetic individual with autonomic neuropathy. He no longer has the capacity to adjust blood flow and respond to local tissue needs; hence, this protection feature is lost. This interference with regulatory power limits the ability to maintain homeostasis. Altered responses of such patients to physiologic, psychologic, and pharmacologic stimuli may lead to severe clinical problems. For example, on exercise, diabetic autonomic neuropathy results in an impaired increase in heart rate and lower blood pressure and aerobic capacity thus significantly diminishing tolerable work loads (46).

It seems reasonable to suspect that, with the development of vascular disease, otherwise relatively asymptomatic autonomic neuropathic defects may play a role in producing transient ischemia or even infarction. Thus they may accelerate, precipitate or act as a modifying determinant of vascular pathologic conditions. In the brain, this could lead to cerebral ischemia and cerebral infarction; in the heart, arrhythmias and conduction defects can lead to cardiac arrest and myocardial infarction; and in the foot, gangrene may result.

There is evidence to suggest that such indeed does take place. In a prospective study, the death rate of diabetic patients with severe autonomic neuropathy was two and one-half times that of those diabetic individuals without comparable autonomic neuropathy (47); thus simple autonomic function tests may prove to be a reliable guide to prognosis. In a study of hospitalized patients with myocardial infarction a two-fold increased mortality in the diabetic versus the nondiabetic patient was observed. This fact could not be correlated with either shock, pulmonary edema, etc., but there was a definite correlation with the presence of arrhythmia and/or conduction defect (48). Arrhythmia and

even cardiac arrest in diabetic patients with advanced autonomic nervous system involvement has been observed (49).

A new dimension has been added by observing variations in the nature of autonomic nerve involvement (50-52). Specifically, it has been shown that the sympathetic and parasympathetic components need not be equally affected. This approach to a chemical dissection affords an opportunity to determine rates and types of progress of these two components of the autonomic nervous system.

This concept could open a new vista in research and suggests a new therapeutic approach to these frustrating problems. It underscores the need for aggressive and intensive research to help elucidate the cause, pathogenesis, treatment and prevention of the neuropathic factors.

Summary

Autonomic nervous system neuropathy has deleterious effects on physiologic functions of the blood vessels and circulation. Altered responses of such patients to physiologic, psychologic and pharmacologic stimuli may lead to serious clinical problems. This in turn may lead to impairment of normal homeostatic mechanisms and interfere with the responses that are necessitated by the continually fluctuating demands for local and distal tissues. These abnormalities superimposed on a pathologic vascular system could play a definitive role in the marked increase in vascular related pathologic conditions in the diabetic patient which is now the leading cause of morbidity and mortality in this disease.

Since male and female diabetic individuals are at equal risk for the development of diabetic neuropathy, this could be one clue to understanding the elimination of the sex difference in the frequency of these complications in the diabetic patients. It might be added, parenthetically, that to date there are no other clues to explain the equalization of risk for both sexes for arteriosclerotic complications in diabetes.

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Students' and Physicians' Knowledge of Hospital Charges at Mount Sinai Hospital and School of Medicine

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Abstract

This study examines knowledge of health care costs among medical students, house officers, and attending physicians in various departments of The Mount Sinai Hospital (MSH) and Mount Sinai School of Medicine (MSSM). Medical personnel, deficient in the basic knowledge of hospital costs and charges, are a major component of the escalating cost of health care in the United States. We measured knowledge of the prices charged by The Mount Sinai Hospital for a variety of inpatient laboratory tests, procedures, and other medical services. The impact of previous cost education and experience within the medical field on knowledge of charges were analyzed. Utilizing pass/fail criteria, and measuring those who failed to meet these criteria, as under or overestimators, we examined responses to twenty-seven queries on specific hospital charges. Of the 280 medical students, house officers, and attending physicians who returned the questionnaire (70% return rate), only 12.5% met the passing criteria and exhibited good knowledge; of those who exhibited poor knowledge, 87.8% were underestimators of the charges. Therefore, neither previous cost education nor experience resulted in increased awareness of charges by medical personnel.

Introduction

The upward spiraling cost of medical care in the United States has become a topic of great concern among economists, governmental agencies, and consumers during the past ten years.

"Between 1965 and 1970, while the cost of living rose 23%, the total cost of hospitalization in the U.S. rose 126%: dietary and pharmacy, 50%; housekeeping, 100%; house staff salaries and education, 130%; and laboratory costs, 229%."

(1)

This unexampled increase in medical care expenditures rose unabated, at a rate of 50% greater than the remainder of the economy in 1974 (2). Clearly, this trend cannot continue without a detrimental effect on both health care and the economy.

In 1977, third-party payers represented 67% of all health care expenditures and 90% of all hospital

expenditures (3). With the three major third-party payers contributing eighty-one cents (Medicare thirty-three cents, Medicaid sixteen cents, and Blue Cross thirty-two cents) (1), out of every inpatient dollar reimbursed to Mount Sinai it is clear that either governmental pressure through legislation or coercion or selective decreased reimbursement rates by the third-party payers will be brought to bear on the medical industry. An attempt to contain these costs through peer group review of medical practices for diagnosis and treatment was attempted with the passage of the Professional Standards Review Organization (PSRO) legislation in 1972 (2). The PSRO Boards try to curb the increase in costs by disallowing repayment for "unjustified" procedures or tests, and by making the hospitals file for certificates of need prior to capital investment. However, the hospitals shift the incurred deficits to other services and the costs continue to rise. Ultimately, the real cost of decreased medical service caused by governmental interference will be borne by society as a whole.

One of the largest components of increased hospital costs has been the rapid growth in both the volume and variety of laboratory tests. The begin-

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ning of this trend occurred over forty years ago. In the ten-year period commencing with 1938, there was a 66 $\frac{2}{3}$ % increase in the number of tests per hospital day. This trend continued until the 1958 figure was twice that of 1938 (4). Since then, an annual increase of 10% in the average number of laboratory (lab) procedures has been reported (4). A somewhat limited AMA study in 1964 showed a rise in the use of the clinical laboratory with an increase in the average number of lab procedures per admission studied—from 3.19 in 1946 to 6.36 in 1961 (4), while other studies have shown the laboratory in teaching hospitals to be a major component in hospital cost (5). Indeed, this may be illustrated at MSH where the number of tests performed in the chemistry laboratories increased from 1.2 million in 1970 to 2.2 million in 1975. The estimated figure for 1977 was 3 million (6). At MSH, ancillary costs (operating room, lab tests, drugs, therapies and medical supplies) contributed 42.4% (\$118.82) in 1977 to the average inpatient cost per day (\$280.55) (1). Therefore, the onus is on the physician, as best qualified, to determine which laboratory tests and procedures are necessary to insure a diagnosis, to recommend and to provide treatment, and to furnish proper follow-up. Thus, it is critical that the physician understand the field of health economics, including the price, cost (7), and benefit of each test or procedure ordered.

Initially, this project was to measure attending physicians', house officers' and medical students' knowledge of the costs of certain laboratory tests and procedures as well as other hospital costs and examine how various factors (experience and education) contribute to this knowledge. Unfortunately, MSH has not developed a list of the true costs per laboratory test or procedure; therefore, this paper measures knowledge of hospital charges.

Charges consist of the actual total cost of the test, procedure, or service plus a percentage of the hospital deficit and are determined by complex formulations (1). At all times, charges are greater than costs. The hospital's deficit is incurred under the third-party payment system which reimburses the hospital on a flat per-diem rate per bed filled at midnight, no matter the number of tests or procedures performed. It was estimated that, in 1977, eighty-four percent of the inpatient population at MSH was covered by one of the three major third-party payers (Medicare, Medicaid, or Blue Cross). However, they represented only eighty-one percent of the inpatient revenues. The 1977 average cost per patient day at MSH was \$280.55 while the reimbursement per patient day was \$261.54 for

Medicare, \$242.69 for Medicaid, and \$246.16 for Blue Cross. Only the remaining sixteen percent of the inpatient population (thirteen percent covered by private insurance and three percent true self-payers) were forced to pay charges which represented nineteen percent of the inpatient revenues (1).

Background

The American Medical Association (AMA) has recently recommended that medical faculty and physicians become more familiar with costs and the utilization of hospital ancillary services (7). This requires that physicians become knowledgeable in all of the many aspects of patient-related medical expenses, including hospital charges. The recommendations echoed by the AMA are not new, but have been voiced for years by both hospital administrators and physicians, as a portion of the high cost of hospital care rests with the physician (8). Indeed, hospital admission itself generates a large number of laboratory tests and therefore costs. The Joint Commission on Hospital Accreditation suggested a battery of required laboratory tests in addition to the admission history and physical examination (9). Typically, this consists of a complete blood count, SMA-6, SMA-12, serological test for syphilis, urinalysis, chest x-ray and electrocardiogram.

The information gained from this routine screening has proved beneficial. It has been found that 3.6% of all routine chest x-rays were positive for disease. Moreover, 11% of routine abdominal films yielded diagnostic information previously overlooked in the history and physical examination (10). Furthermore, routine blood analyses were significant in 38% of the patients and, in 75% of these cases, the ultimate diagnosis was directly aided or affected by this previously unexpected information (11). However, in recent years with the proliferation of new and more sensitive laboratory tests, concern has been voiced that these procedures are being abused (9, 12, 13). Reliance on these tests may reflect the physicians' inexperience in obtaining an adequate history or physical examination. In ordering a "shotgun" approach or a large battery of laboratory tests, the physician is able to partially compensate for his deficiencies. Other uses of laboratory tests that tend to increase hospital costs include "roundsmanship" (the desire to impress peers) or protect against litigation (9).

The impact of laboratory tests on the improvement of health care delivery was examined in a study at the University of Rochester. Utilizing

matched groups of patients with diabetic ketoacidosis, there was a 27% increase in laboratory tests, from 1966 to 1969; however, the outcome of care measured by the length of hospitalization was not significantly different between the two groups (5). As part of the same study, all laboratory tests were performed as ordered, but certain results were not reported to the requesting physicians. Not a single request for the results was made, thereby illustrating a superfluous ordering of procedures (5).

A retrospective analysis at the Durham Veterans Administration Hospital concluded that only 5% of the laboratory results were significant in diagnosis and treatment. Through education of the house-staff, a 25% reduction in laboratory tests was obtained without a significant loss of essential diagnostic or therapeutic information (14).

The question that then arises is whether physicians and medical students are cognizant of the costs of laboratory tests they so frequently request. Studies have shown that medical personnel have a poor fund of knowledge and the tendency to underestimate laboratory costs (2, 15). One must also consider those factors difficult to quantify, that is, the danger and/or discomfort imposed upon the patient in the performance of tests and procedures. Examples include hypersensitivity reactions to contrast material, perforation of a viscus, loss of a pulse, etc. It is difficult to draw a sharp line between tests and procedures that are decisive and indecisive. Does the laboratory test yield new information or is it overkill? The decision regarding appropriate tests must be left to the individual physician who is guided by the patient's best interests.

Materials and Methods

An anonymous, fill in type of questionnaire was constructed and pretested, querying medical students, house officers and attending physicians in various departments of MSH and the School, to estimate the prices charged at MSH for twenty-five common inpatient laboratory tests (2), procedures, or materials, the minimum per-diem price of a MSH bed in their specialty, and the price of any MSH ICU bed. The individual tests and procedures were chosen since they represent frequently utilized tools in the diagnosis and treatment of hospitalized patients.

The questionnaire was personally, randomly distributed and collected, by the authors, at conferences, student lectures, and through interdepartmental mail. In all, 400 questionnaires were distributed and 291 (72.75%) were returned. Of these, 11 were returned incomplete and were discarded. This

paper is based on the completed 280 questionnaires, or 70% of the total distributed.

The effect of two major variables on the knowledge of charges were examined: the impact of previous cost or health economics education, or lectures, and experience within the medical profession. The former, for simplicity, to be further denoted as cost education or education, occurred if the individual claimed to have attended a lecture in health economics and/or in which the cost of laboratory tests or procedures was discussed. The latter variable, experience, was examined by division of the data into that from medical students, house officers, and attendings. The medical students were separated into two major groups: first and second (MSSM 1 + 2) and third (MSSM 3) year students. The third year students were further divided into groups that spent all three years at Sinai—two basic science years and one clinical year—(MSSM 3-3), those that transferred to Sinai after two years of basic sciences (MSSM 3-1), and those who neglected to note the number of years associated with Mount Sinai (MSSM 3-?). The house officers were grouped as post graduate year (PGY) 1 (interns), PGY 2, PGY 3, and PGY 4-Fellows while the attendings were separated into nonpsychiatric and psychiatric attending physicians. Experience in the medical profession and to a given institution was measured as the number of years in practice and the number of patients hospitalized at that institution (exposure). The former group of attendings were subdivided into four smaller groups:

Group	Years in Practice	No. of Patients Hospitalized at MSH in the Past 12 Months
1	≤10	≤20
2	≤10	>20
3	>10	≤20
4	>10	>20

The psychiatrists, because of the small number of patients that they hospitalize, were only subdivided into those practicing ten years or less and those practicing greater than ten years.

Following an examination of the data and utilizing a study by Skipper, et al (2) we considered a correct answer as the actual price charged $\pm 25\%$. These rates were calculated from the charges listed in the *MSH Pricing Catalogue* of June 28, 1978 (16) and are listed in Table I. After a further review of the data, we arbitrarily set a passing grade of eleven out of the remaining twenty-seven questions (40.74%), since a higher passing grade would have resulted in a significant decrease in the percentage passing (Table II). The individuals who failed to

meet the passing criteria of eleven correct responses were evaluated as over or under estimators, and those who equally over and under estimated were discarded. The remaining questionnaires were evaluated for the existence of previous cost education and experience level of training.

Throughout the analysis, except where specifically mentioned, psychiatric and nonpsychiatric attending physicians were considered separately. This

TABLE I

*The Actual Prices Charged at MSH for Various Laboratory Tests and Procedures and the Normal Range (Price $\pm$ 25%)**

Test or Procedure	Actual Price Charged	Normal Range**
1. CBC	\$ 31.00	\$ 23.25-38.75
2. CBC with differential	41.00	30.75-51.25
3. Hematocrit	11.00	8.25-13.75
4. One unit of whole blood (450 ml)	34.50	25.88-43.13
5. One unit of packed red blood cells (200 ml)	34.50	25.88-43.13
6. SMA-6	47.00	35.25-58.75
7. SMA-12	85.00	63.75-106.25
8. Serum potassium	30.00	22.50-37.50
9. Serum creatinine	30.00	22.50-37.50
10. Blood glucose	30.00	22.50-37.50
11. Prothrombin time	25.00	18.75-31.26
12. Arterial blood gas (ABG)	87.00	65.25-108.75
13. ESR	30.00	22.50-37.50
14. Urinalysis	13.00	9.75-16.25
15. Urine culture and sensitivity (C&S)	61.00	45.75-76.25
16. Routine EKG	58.50	43.88-73.13
17. Echocardiogram	59.00	44.25-73.75
18. Chest x-ray (PA and lateral)	79.00	59.25-98.75
19. Portable chest x-ray (AP)	96.00	72.00-120.00
20. Abdominal flat plate (KUB)	57.00	42.75-71.25
21. UGI series with small bowel follow-through	302.00	226.50-377.50
22. Barium enema	189.00	141.75-236.25
23. IVP (6 films, nontimed)	129.00	96.75-161.25
24. Abdominal sonography hour operating room time	328.00	246.00-410.00
26. Minimum daily price of a MSH bed in your specialty		
a. nonpsychiatry	230.00	172.50-287.50
b. psychiatry	265.00	198.75-331.25
27. The price per day of any MSH ICU bed	430.00	322.50-537.50

* Based on The Mount Sinai Hospital (MSH) Pricing Catalog, computer printout of June 28, 1978, as referenced in paper.

** Range = (actual price - 25% of actual price) to (actual price + 25% of actual price) or, Range = actual price $\pm$ 25%.

TABLE II

Comparison of Those Passing Utilizing High and Low Criteria

Group	Low Criteria ≥ 11 Correct Answers	High Criteria ≥ 14 Correct Answers
Medical Students	13	3
House Officers	10	0
Nonpsychiatric Attending	9	2
Psychiatrists	3	2
Total	35	7

was suggested by members of the Department of Psychiatry, and instituted, as not to skew the data, because psychiatric attendings rarely admit patients and are less exposed to hospital laboratory tests and/or procedures. Finally, in the analysis of the data, significance was either measured by Chi-Square or the Fischer Exact Probability Test.

Discussion

The purpose of this study was to examine the knowledge of health care professionals (medical students, housestaff, and attending physicians) at The Mount Sinai Hospital and Mount Sinai School of Medicine of the charges of frequently ordered and utilized laboratory tests, procedures, and medical services. Our contention was that one should have increased knowledge with either cost education (in the form of lectures on cost or health economics) and/or increased experience in the medical profession. Therefore, we analyzed both cost education and experience.

Of the 280 returned questionnaires 85 were from medical students, 70 housestaff, 79 nonpsychiatric attending physicians and 46 psychiatrists. 116 of the respondents claimed previous cost education (54.1% of the medical students, 31.4% of the housestaff, 49.4% of the nonpsychiatric attendings and 19.6% of the psychiatrists), while 164 did not.

Overall, only 12.5% of those surveyed were able to fulfill the original arbitrary criteria for passing and thus exhibit good knowledge of the charges (Table III). If the passing criteria were increased from 40.74% to 51.8% (fourteen correct responses), only 20% of those with good knowledge were able to meet the higher criteria. Therefore, to increase the number of individuals in the passing category, we utilized the lower passing criteria. If one excludes the psychiatrists, those passing increased to 13.7%. Evaluating the data, attending physicians, house staff and medical students have a limited knowledge of the charges of laboratory tests, procedures and

TABLE III
Pass-Fail*
in Actual Numbers (and as Percentages)

Group	Total			With Cost Education			Without Cost Education		
	Pass	Fail	Total	Pass	Fail	Total	Pass	Fail	Total
<i>Students:</i>									
MSSM 1+2	0 (0)	7 (100)	7	0 (0)	5 (100)	5 (71.4)	0 (0)	2 (100)	2 (28.6)
MSSM 3-1	3 (15)	17 (85)	20	3 (27.3)	8 (72.7)	11 (55)	0 (0)	9 (100)	9 (45)
MSSM 3-3	9 (20.5)	35 (79.5)	44	6 (26.1)	17 (73.9)	23 (52.3)	3 (14.3)	18 (85.7)	21 (47.7)
MSSM 3-?	1 (7.1)	13 (92.9)	14	1 (14.3)	6 (85.7)	7 (50)	0 (0)	7 (100)	7 (50)
Total	13 (15.3)	72 (84.7)	85 (30.4)	10 (21.7)	36 (78.3)	46 (54.1)	3 (7.7)	36 (92.3)	39 (45.9)
<i>House Staff:</i>									
PGY-1	2 (12.5)	14 (87.5)	16	1 (14.3)	6 (85.7)	7 (43.8)	1 (11.1)	8 (88.9)	9 (56.2)
PGY-2	2 (10.5)	17 (89.5)	19	1 (16.7)	5 (83.3)	6 (31.6)	1 (7.7)	12 (92.3)	13 (68.4)
PGY-3	2 (15.4)	11 (84.6)	13	0 (0)	4 (100)	4 (30.8)	2 (22.2)	7 (77.8)	9 (69.2)
PGY-4 → Fellow	4 (18.2)	18 (81.8)	22	0 (0)	5 (100)	5 (22.7)	4 (23.5)	13 (76.5)	17 (77.3)
Total	10 (14.3)	60 (85.7)	70 (25)	2 (9.1)	20 (90.9)	22 (31.4)	8 (16.7)	40 (83.3)	48 (68.6)
<i>Non-Ψ</i>									
<i>Attendings:</i>									
≤10 yrs, ≤20 pts	1 (6.7)	14 (93.3)	15	1 (12.5)	7 (87.5)	8 (53.3)	0 (0)	7 (100)	7 (46.7)
≤10 yrs, >20 pts	2 (11.8)	15 (88.2)	17	1 (14.3)	6 (85.7)	7 (41.2)	1 (10)	9 (90)	10 (58.8)
>10 yrs, ≤ 20 pts	1 (5.6)	17 (94.4)	18	1 (11.1)	8 (88.9)	9 (50)	0 (0)	9 (100)	9 (50)
>10 yrs, >20 pts	5 (17.2)	24 (82.8)	29	3 (20)	12 (80)	15 (51.7)	2 (14.3)	12 (85.7)	14 (48.3)
Total	9 (11.4)	70 (88.6)	79 (28.2)	6 (15.4)	33 (84.6)	39 (49.4)	3 (7.5)	37 (92.5)	40 (50.6)
<i>Ψ Attendings:</i>									
≤10 Yrs	0 (0)	14 (100)	14	0 (0)	5 (100)	5 (35.7)	0 (0)	9 (100)	9 (64.3)
>10 Yrs	3 (9.4)	29 (90.6)	32	0 (0)	4 (100)	4 (12.5)	3 (10.7)	25 (89.3)	28 (87.5)
Total	3 (6.5)	43 (93.5)	46 (16.4)	0 (0)	9 (100)	9 (19.6)	3 (8.1)	34 (91.9)	37 (80.4)
Grand Total									
̄ Ψ Attendings	32 (13.7)	202 (86.3)	234	18 (16.8)	89 (83.2)	107 (45.7)	14 (11.0)	113 (89)	127 (54.3)
Grand Total									
̄ Ψ Attendings	35 (12.5)	245 (87.5)	280 (100)	18 (15.5)	98 (84.5)	116 (41.4)	17 (10.4)	147 (89.6)	164 (58.6)

* Ψ denotes psychiatry

MSSM = Mount Sinai School of Medicine

PGY = Postgraduate year

medical services. An analysis of the sample that indicated previous cost education resulted in a 3% increase in the percentage passing over the total; while, there was a 5.1% increase in the percentage passing when comparing those with and without cost education.

Of those who demonstrated poor knowledge and failed to meet the criteria for passing, 87.8% were underestimators and 7.3% overestimators (Table IV). The remaining 4.9% had an equal number of over and underestimates. Excluding the psychiatrists, 88.6% were underestimators, 7.9% were overestimators, and 3.5% had an equal number of over and underestimates. Therefore, the tendency for the total sample was to underestimate the charges. This corresponds with the conclusions reached by Skipper, et al (2). Furthermore, when comparing medical students, housestaff and attending physicians, there was also a greater tendency to under-

estimate with increased experience, or time spent, in the medical profession ($p < 0.05$).

Examining the variable experience, there was no increase in those with good knowledge among medical students, housestaff, and nonpsychiatric attending physicians (15.3%, 14.3%, and 11.4%, respectively). An increasing number of medical students exhibited good knowledge of costs with increased grade level. However, these values were found not to be statistically significant. Moreover, there was no significant difference between the third year medical student and intern groups, indicating that two years of increased clinical exposure did not add to a greater knowledge of costs.

Although there was a propensity for increased knowledge among housestaff with more years of training, this was not found to be statistically significant. This trend remained unchanged even when analyzing the group with cost education. Nonpsy-

TABLE IV
Over vs Under Estimators*
in Actual Numbers (and as Percentages)

Group	Failed				With Cost Education			Without Cost Education		
	Total	Over- estimators	Under- estimators	Over Equal Under Estimates	Over- estimators	Under- estimators	Total	Over- estimators	Under- estimators	Total
<i>Students:</i>										
MSSM 1+2	7	0 (0)	7 (100)	0 (0)	0 (0)	5 (100)	5 (71.4)	0 (0)	2 (100)	2 (28.6)
MSSM 3-1	17	3 (17.6)	14 (82.4)	0 (0)	2 (25)	6 (75)	8 (47.1)	1 (11.1)	8 (88.9)	9 (52.9)
MSSM 3-3	35	4 (11.4)	30 (85.7)	1 (2.8)	1 (5.9)	16 (94.1)	17 (48.6)	3 (17.6)	14 (82.4)	17 (48.6)
MSSM 3-?	13	1 (7.7)	12 (92.3)	0 (0)	1 (16.7)	5 (83.3)	6 (46.2)	0 (0)	7 (100)	7 (53.8)
Total	72	8 (11.1)	63 (87.5)	1 (1.4)	4 (11.1)	32 (88.9)	36 (50)	4 (11.4)	31 (88.6)	35 (48.6)
<i>House Staff:</i>										
PGY-1	14	3 (21.4)	10 (71.4)	1 (7.1)	2 (33.3)	4 (66.7)	6 (42.9)	1 (14.3)	6 (85.7)	7 (50)
PGY-2	17	1 (5.9)	15 (88.2)	1 (5.9)	0 (0)	5 (100)	5 (29.4)	1 (9.1)	10 (90.9)	11 (64.7)
PGY-3	11	2 (18.2)	9 (81.8)	0 (0)	1 (25)	3 (75)	4 (36.4)	1 (14.3)	6 (85.7)	7 (63.6)
PGY4 → Fellow	18	1 (5.6)	16 (88.9)	1 (5.6)	0 (0)	5 (100)	5 (27.8)	1 (8.3)	11 (91.7)	12 (66.7)
Total	60	7 (11.7)	50 (83.3)	3 (5)	3 (15)	17 (85)	20 (33.3)	4 (10.8)	33 (89.2)	37 (61.7)
<i>Non-Ψ</i>										
<i>Attendings:</i>										
≤10 yrs, ≤20 pts.	14	0 (0)	14 (100)	0 (0)	0 (0)	7 (100)	7 (50)	0 (0)	7 (100)	7 (50)
≤10 yrs, >20 pts.	15	0 (0)	14 (93.3)	1 (6.7)	0 (0)	5 (100)	5 (33.3)	0 (0)	9 (100)	9 (60)
>10 yrs, ≤20 pts.	17	1 (5.9)	15 (88.2)	1 (5.9)	1 (12.5)	7 (87.5)	8 (47.1)	0 (0)	8 (100)	8 (47.1)
>10 yrs, >20 pts.	24	0 (0)	23 (95.8)	1 (4.2)	0 (0)	11 (100)	11 (45.8)	0 (0)	12 (100)	12 (50)
Total	70	1 (1.4)	66 (94.3)	3 (4.3)	1 (3.2)	30 (96.8)	31 (44.3)	0 (0)	36 (100)	36 (51.4)
<i>Ψ Attendings:</i>										
≤10 yrs	14	1 (7.1)	13 (92.9)	0 (0)	1 (20)	4 (80)	5 (35.7)	0 (0)	9 (100)	9 (64.3)
>10 yrs	29	1 (3.4)	23 (79.3)	5 (17.2)	0 (0)	1 (100)	1 (3.4)	1 (4.3)	22 (95.7)	23 (79.3)
Total	43	2 (4.7)	36 (83.7)	5 (11.6)	1 (16.7)	5 (83.3)	6 (14.0)	1 (3.1)	31 (96.9)	32 (74.4)
Grand Total										
̄ Ψ Attendings	202	16 (7.9)	179 (88.6)	7 (3.5)	8 (9.2)	79 (90.8)	87 (43.1)	8 (7.4)	100 (92.6)	108 (53.5)
Grand Total										
̄ Ψ Attendings	245	18 (7.3)	215 (87.8)	12 (4.9)	9 (9.7)	84 (90.3)	93 (38.0)	9 (6.4)	131 (93.6)	140 (57.1)

* Ψ Denotes Psychiatry

MSSM = Mount Sinai School of Medicine

PGY = Postgraduate year

chiatric attending physicians also demonstrated a higher percentage passing with increased years in practice (9.4% for those practicing less than or equal to ten years and 12.8% for those practicing greater than ten years). Another component of increased experience at a given institution is the number of patients hospitalized by a physician within any given time period. The data demonstrated that there was no statistically significant difference between these groups (6.1% passing with less than or equal to twenty patients hospitalized per year versus 15.2% for those with greater than twenty patients hospitalized per year). Finally, there was no statistical difference between nonpsychiatric attending physicians and psychiatrists who demonstrated good knowledge. In summary, there was no statistical significance for increased knowledge of

costs with increased experience in the medical profession.

The number of individuals claiming previous cost education differed among the various experience groups, with 54.1% of medical students, 31.4% of housestaff, and 49.4% of the non psychiatric attending physicians answering affirmatively. Medical students with cost education exhibited an increased percentage passing over students without cost education: 21.7% and 7.7% respectively ($p < 0.10$).

Paradoxically, almost twice the percentage of housestaff without education (16.7%) demonstrated good knowledge as compared to those with cost education (9.1%). This value was not found to be statistically significant and this may have been a function of the small sample size. The same was true for the psychiatrists of whom 8.1% without cost

education and 0% with cost education passed. The aforementioned paradoxical results were reversed among the nonpsychiatric attending physicians, 15.4% of whom with cost education passed as compared to 7.5% without cost education. In summary, the global effect of cost education was to increase the percentage of those passing. However, when individual experience groups were examined this relation did not hold. Possible explanations are that different groups received varying types (quality and quantity) of cost education and the temporal relation of this education with the level of training.

Conclusions

The entrance of third-party payers into the health care profession has distorted the delicate balance of a free market economy. The majority of patients hospitalized in the United States are covered by third-party payers and only indirectly bear the costs of their medical care; the aggregate costs are collectively borne by society as insurance premiums and taxes.

We examined physicians', house officers', and medical students' knowledge of the charges for basic laboratory tests, procedures and medical services at MSH in 1978. Our belief was that one of the components of skyrocketing health care costs was that medical personnel lack basic knowledge of hospital charges. Of those surveyed, only 12.5% demonstrated good knowledge and of those exhibiting poor knowledge, 87.8% were underestimators of charges. A second hypothesis was that education and/or experience in the medical profession would lead to increased knowledge of these charges. We found that neither previous cost education nor experience led to an increased awareness of hospital prices. Whether or not this is indicative of the quality of the education being offered or the temporal relationship between the education received and level of training must be left to future studies.

As previously discussed, it is possible to reduce the number of laboratory tests and procedures without adversely affecting patient care or outcome. We propose that The Mount Sinai Hospital and School of Medicine institute a program of cost awareness among attendings, housestaff and medical students. Moreover, this program should be part of a broadened medical school curriculum that incorporates cost/benefit analysis as well as the indications for, risks and interpretation of laboratory tests and other medical procedures.

The escalating cost to society of medical care must be contained before the government is forced

to actively take over the health care industry. The days in which government funds flowed into the medical sector without restriction are over. There is no such thing as a free lunch and the people—via the government—pay a large proportion of the hospital bill and demand efficiency. Hopefully, the day will not come when doctors are forced, by nonmedical personnel, to curtail their diagnostic and therapeutic abilities because certain diagnostic tests or therapeutic regimens are too financially costly. With the cost of laboratory tests contributing a large share to the escalating cost of medical care it is important, if not imperative, that physicians understand the cost of laboratory tests, procedures and medical care, not so they will curtail vital services, but rather that they will understand that the best short-run solution is the elimination of unnecessary tests and procedures.

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Ventriculitis Complicating Gram Negative Meningitis in an Adult. Diagnosis by Radioisotope Brain Scanning and Computerized Cranial Tomography

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Abstract

A safe, noninvasive diagnostic modality to determine the existence of ventriculitis complicating gram negative meningitis in adults would help define the natural history and therapy of this disease. In this report, a patient with *Enterobacter* meningitis is described in whom the diagnosis of ventriculitis was first suspected by radionuclide brain scanning and computerized cranial tomography and later confirmed by histologic examination. Studies are needed to assess sensitivity and to determine the predictive value of serial scanning during antimicrobial therapy.

Introduction

The incidence and significance of ventriculitis complicating gram negative bacillary meningitis in adults is unknown. This may in part be attributed to lack of a safe noninvasive procedure for diagnosis of ventricular infection. In this communication we report the successful use of radionuclide brain imaging and cranial computerized tomography in the diagnosis of ventriculitis complicating gram negative meningitis in an adult. The mechanism of visualization is discussed.

Case Report

A 54-year-old male was admitted to the Veterans Administration Medical Center, Bronx, New York for evaluation of a right hilar mass discovered on chest x-ray. Three days after a normal fiberoptic bronchoscopy, the patient developed a right lower

lobe pneumonia with bacteremia due to *Enterobacter sp.* Clinical improvement and roentgenologic resolution occurred during a two-week course of carbenicillin and gentamicin but fever persisted. Three days after antibiotics were discontinued the patient became lethargic and developed nuchal rigidity. Cerebrospinal fluid (CSF) examination showed a protein of 162 mg/dl, a glucose of 3 mg/dl and 12,700 white cells/mm³, 98% polymorphonuclears. Culture of blood and cerebral spinal fluid (CSF) grew *Enterobacter sp.* with biochemical profile and antibiogram identical to that of the earlier isolates from blood and sputum. Chloramphenicol was begun intravenously but, after four days, the organism developed resistance. Carbenicillin at 40 gms/day intravenously, trimethoprim-sulfamethoxazole at 640 mg/3200 mg per day orally and gentamicin 240 mg/day intravenously were substituted. In addition, 8 mg gentamicin was given intrathecally by lumbar administration every 24 hours. Corticosteroids were used for control of possible cerebral edema. CSF culture became negative after three days and the patient slowly improved.

During the first week of therapy, a brain scan obtained two hours after administration of 20 mCi of 99m technetium-pertechnetate diethylenetri-

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minepentaacetic acid (Tc-DTPA) revealed increased activity bilaterally in deep parietal foci corresponding to the region of the posterior horns of the lateral ventricles (Fig. 1). Ventricular aspiration was deferred because of clinical improvement. Intrathecal gentamicin was discontinued after three weeks, and carbenicillin and parenteral gentamicin after seven weeks of therapy. Repeat brain scans continued to demonstrate ventricular uptake, but progressively less so. Computerized cranial tomography (CT scan) obtained during the fifth week of therapy showed ventricular enlargement with enhancement of the walls of the third and lateral ventricles after administration of an iodinated contrast agent (Fig. 2).

As trimethoprim-sulfamethoxazole was well tolerated, it was elected to continue this medication until the abnormal ventricular uptake on brain scan



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FIG. 1. Posterior and right lateral brain scintiscan views reveal abnormal bilateral lateral ventricular foci.

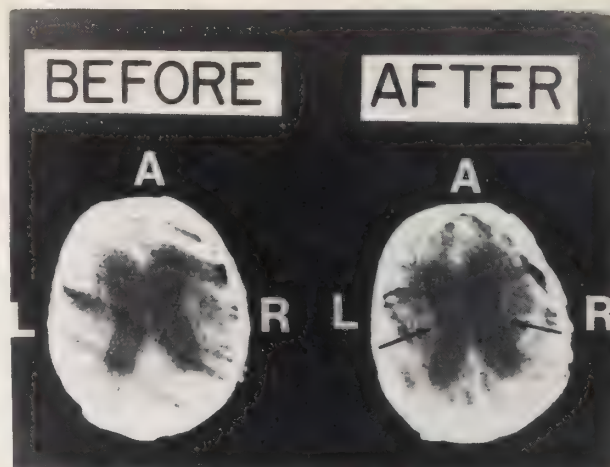


FIG. 2. Precontrast computed cranial tomogram reveals dilated lateral ventricles. Postcontrast image additionally demonstrates a ventricular "halo" of increased periventricular attenuation compatible with ventriculitis.

disappeared. The patient also received a brief course of isoniazid and rifampin because of an equivocally positive acid fast smear on sputum and the unexplained right hilar mass. Mycobacterial cultures on sputum and CSF however eventually proved negative. (The *Enterobacter sp* was resistant to rifampin by disc agar diffusion.)

Although neurologic status remained stable, the patient was left with a dense organic mental syndrome, he continued to lose weight. Repeat CSF examinations persistently showed 20 to 30 white cells with approximately 30% polymorphonuclears. Glucose and protein values were within normal limits and cultures were negative. On the 127th hospital day, the patient aspirated and expired.

Histologic examination of the occipital horns of both lateral ventricles revealed granulation tissue with an inflammatory reaction composed of mononuclear cells, histiocytes, and extensive cellular debris (Fig. 3). Less marked but similar chronic inflammatory changes were present in the leptomeninges. Fungal, acid fast and bacterial stains failed to demonstrate organisms; unfortunately no postmortem cultures were obtained. Other findings included a peripheral adenocarcinoma of the right lung metastatic to right hilar nodes, gall bladder and adrenals. There was no evidence of tumor cells in meninges or brain.

Discussion

The mortality of gram negative bacillary meningitis in adults approaches 50%, and remains high even with combined parenteral and intralumbar

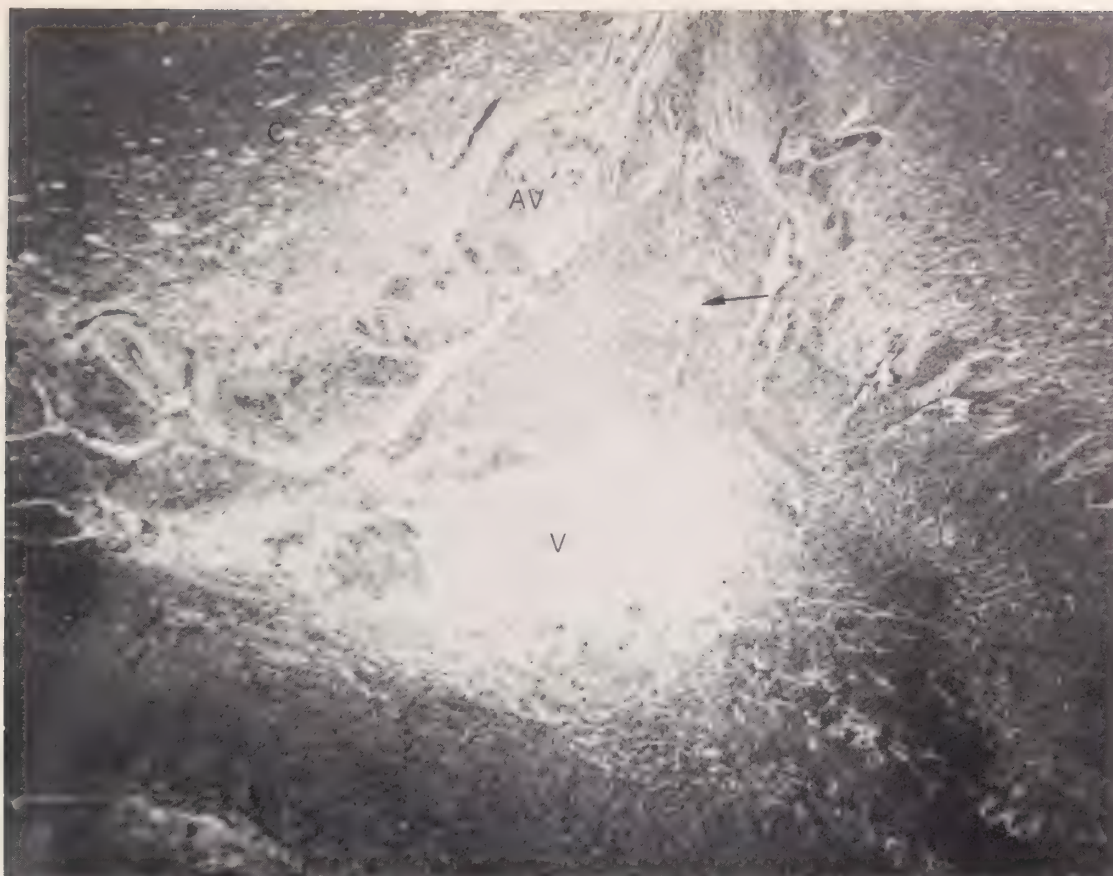


FIG. 3. Postmortem histologic confirmation of ventriculitis (AV-arachnoidal villi, C-cortex, V-occipital horn of lateral ventricle, Arrow-inflammatory exudate within the ventricle). 20×

intrathecal aminoglycoside therapy (1-3). An explanation suggested to account for some therapeutic failures is inadequate treatment of unsuspected ventriculitis (2, 4). It is well documented that parenteral or parenteral plus intralumbar aminoglycoside administration does not achieve antibacterial drug levels in the ventricles, whereas intraventricular therapy can deliver such concentrations throughout the subarachnoid space (4).

However, neither the incidence nor the significance of ventriculitis has been ascertained in adults, as the diagnosis is rarely made antemortem. There has been understandable reluctance to perform ventricular aspiration routinely, the recognized diagnostic modality, because it requires movement of the patient to a neurosurgical operating room for burr holes and because of potential morbidity including spread of infection, intracranial hemorrhage and even death (5, 6).

A safe, noninvasive method for detection of ventriculitis would be highly desirable. Ventricular uptake on radionuclide brain scintiscanning was first recognized as indicative of ventriculitis by Deland

and Wagner using ^{99m}Tc -pertechnetate in an adult with miliary tuberculosis (7). Visualization with conventional radionuclide brain scintiscanning in ventriculitis is presumably related to a breakdown in the blood brain barrier in the ependymal membranes and periventricular portion of the brain induced by the adjacent inflammatory process (8-13). In part, visualization may be related to a breakdown in the blood-ventricular fluid barrier. Assay of radioactivity on ventricular fluid was positive in the one instance in which it was performed (8). Uptake may be enhanced with time. Optimal visualization may occur four or more hours after injection of the radionuclide (10). Resolution of the inflammatory process has been associated with improvement in the scan findings and completely normal scans have been seen in follow up (8). How closely the temporal course of resolution of scan findings parallels bacteriologic cure is unknown.

Ventricular uptake is not a normal variation and is rarely seen in conditions other than ventriculitis. The ^{99m}Tc -pertechnetate normally secreted by choroid plexus will lead to visualization of the choroid

in approximately 15% of patients. However, this excretion is routinely blocked by administration of perchlorate (14). The newer chelated radiopharmaceuticals (eg. tc-DTPA) are not excreted by choroid and do not require perchlorate blockage. Failure of perchlorate administration or effect is suggested by uptake in salivary glands in which secretion of technetium is similar to choroid. Fulmer and Sfakianakis after perchlorate administration, observed cerebral ventricular visualization in only four of 1200 ^{99m}Tc -pertechnetate brain scans performed on children and in none of 12,500 scans on adults. Three of the four children with positive scans had ventricular infection and the last had hyperthermic or anoxic brain injury (8). Other than with infection, visualization in the ventricular system has been seen in patients with choroid plexus papillomas and in children following intrathecal (intralumbar) methotrexate (15).

Ventriculitis can also be diagnosed by cranial computed tomography as illustrated by our patient and other reports (16-18). Localized ependymal enhancement following administration of intravenous contrast media is the usual finding, presumably related to localized disruption of the blood brain barrier. Pus located within the ventricles may appear with higher density than CSF. In one patient, the numerical output of computerized tomography could be correlated with ventricular fluid protein concentrations (18).

At present, the sensitivity of radionuclide and CT scanning for the detection of ventriculitis has not been defined clinically or in an animal model. Nor are there data with which to compare these diagnostic modalities for either sensitivity or specificity. Studies designed to obtain this information deserve consideration.

In summary radionuclide brain scanning or computed cranial tomography are safe, noninvasive procedures that may be useful for certain patients in the diagnosis of ventriculitis complicating gram negative meningitis. The significance of a normal study in such patients is unknown at the present time. Similarly, more data are required to assess the value of serial scans in following therapy.

Acknowledgment

We wish to thank Miss Carrie Zambrzycki for assistance.

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Dissociated Sensory Loss and Visual Evoked Potentials in a Patient with Pernicious Anemia

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Abstract

A patient with megaloblastic anemia following gastrectomy is described. Neurologic findings included loss of tendon reflexes, mild ataxia, and sensory signs. Remarkably, vibration sense was absent in the lower limbs, but fairly normal in the hands, whereas joint-position sense was absent in the hands, but preserved in the feet. Visual evoked potentials (VEP) showed abnormally prolonged latency. Following institution of B₁₂ therapy there was little change in the patient's sensory status, but VEP latency improved. The basic pathophysiologic processes are discussed.

Introduction

While pernicious anemia (PA) is by definition a hematologic disorder, it often manifests itself as a dysfunction of the nervous system. The spinal cord is the most commonly affected site, but cerebral and ophthalmologic manifestations also occur as well as evidence of peripheral neuropathy (1, 2). It was noted (3) that in some patients disturbed vision and other neurologic symptoms may antedate hematologic abnormalities. The visual manifestations of pernicious anemia commonly are impaired visual acuity and central or cecocentral scotomata. These visual field defects are often bilateral, although not necessarily symmetric, but they can be unilateral. The fundi are conspicuously normal until optic pallor develops late in the course. Apparently visual presentation of PA is rare: in a survey of the literature Hamilton (4) found only a few case histories. Holmes (1) reported three patients: each complained of progressive visual loss for various durations, one of them for twelve months before the

nature of their disease was diagnosed. We describe unusual neurologic and subclinical visual manifestations in a patient who suffered from megaloblastic anemia following gastrectomy. Of special interest in this case are the results of visual evoked potential testing before and after vitamin B₁₂ treatment.

Case Report

This 80-year-old woman was admitted for the chief complaint of numbness of both hands and feet. She was in good general health until eight months prior to admission, when she began to notice progressive impairment of holding and grasping objects in either hand; often she dropped them involuntarily. Her handwriting became less legible, the letters were becoming larger. Three weeks prior to admission, a bandlike sensation appeared around her chest and a bilateral tingling sensation from her fingers up to the elbows and from the toes up to her knees. Eighteen years previously, she had undergone subtotal gastrectomy for an ulcer, and received a course of vitamin B₁₂ treatment for anemia ten years prior to admission. She was told she had arthritis of both knees. On examination, she appeared poorly nourished. She was not oriented in time, could not remember the names of the presidents of the United States, and made errors in reversing five letter words and digits. She could not

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add or subtract two digit numbers. Of three items she forgot all in five minutes. Face-hand test was 1 plus (5). Flow of speech, naming, and sentence repetition were normal. Cranial nerve functions and strength in the upper extremities were intact. There was weakness of dorsiflexion of both feet. Tendon reflexes were absent in the lower limbs, and the plantar responses were flexor. There was some ataxia on finger-to-nose and heel-knee-shin tests. Romberg test was positive. Most sensory modalities were severely affected in the lower part of the body, with a sensory level for vibration and pin-prick at about T6, and graphanesthesia in the lower limbs. However, joint-position sense was close to normal in the toes but markedly impaired in the fingers and wrists. Two-point discrimination was over 20 mm in the fingertips.

Admission laboratory findings were Hgb 4.7; HCT 34.7; RBC 2.76; MCV 126; MCH 42.6; Reticulocytes 1%; WBC, platelets, PTT, ESR normal. Serum iron was 103 $\mu\text{g}/\text{ml}$. TIBC was 262. B_{12} level was 9 $\mu\text{g}/\text{ml}$. SMA 18 was normal. Bone marrow aspirate was interpreted as showing subtle changes of megaloblastosis. VEP testing performed with a checkerboard and with a grating pattern stimulus (6) showed abnormal latency: the major positive deflection occurred at 138 msec in the right eye (O.D.) and 146 msec in the left eye (O.S.). These values are more than 3 S.D. above the normal limits established in previous studies (6). Following treatment with B_{12} intramuscularly, the VEP latencies one month later were 126 msec O.D. and 134 msec O.S. Thus, the latency became shorter by 12 msec in both eyes following therapy. The EP latency

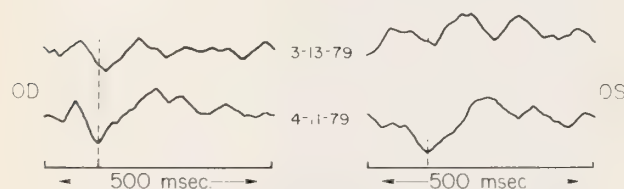


FIG. 1. Visual evoked potentials (VEP) to an alternating vertical grating pattern of 2.3 cycles/degree obtained in the right eye (O.D.) and the left eye (O.S.) of the patient with pernicious anemia. The rate of pattern reversal was 1 Hertz. The timescale is 500 msec. The top trace was obtained before and the bottom trace was obtained following therapy in each eye. The first downgoing hump in both traces is called the major positive wave of the VEP. A vertical line drawn through the evoked potential identifies in the bottom trace of each eye the major positive wave. This occurs in normals with a latency between 100 and 135 milliseconds. Notice from top to bottom trace that in the right eye the latency decreased and the amplitude increased. In the left eye, it is somewhat difficult to identify the major positive wave due to the very low amplitude response. However, it is possible to identify it by comparing it with the trace below, representing the VEP after treatment.

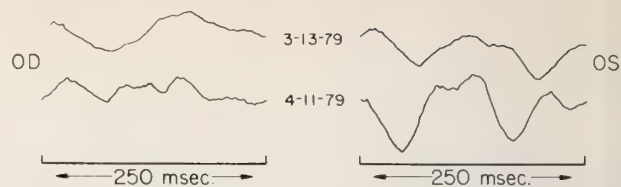


FIG. 2. VEP-s to the same grating pattern as in Fig. 1. This was presented in a pattern on, pattern off mode at the rate of 8 Hertz. EP-s were obtained before and after therapy. The timescale is 250 milliseconds. In the right eye there is little response prior to therapy. Following therapy, spectral analysis of the EP trace identified mostly second harmonic response. In the left eye there is some first harmonic response prior to therapy. Following therapy the apparent amplitude of the response (bottom trace) increased and spectral analysis showed that in comparison to the EP before therapy the amplitude increase was largely due to the now present second harmonic response. Also notice phase shift of the response. For both EP-s the stimulus pattern subtended 4° at the eye and was viewed foveally.

changes in the patient are significant and are presumably due to therapy, as in normal subjects fluctuations of VEP latency have an S.D. of 4 msec. This improvement of the VEP latency is especially remarkable since as a result of therapy, there was less certain change within the first months in the clinical status of the patient. Some observers could detect mild improvement in her ability to respond to questions directed to her, and she detected vibration with greater reliability.

Discussion

Subtotal gastrectomy may lead to the development of vitamin B_{12} deficiency with its hematologic and neurologic manifestations (2). The most common neurologic complication is subacute combined degeneration of the spinal cord which is an anatomical diagnosis, indicating sclerosis (that is, demyelination) of the posterior and lateral columns of the spinal cord. These pathways are probably vulnerable because they contain thickly myelinated fibres. Fibres of that size elsewhere may be similarly affected by B_{12} deficiency, underlying the peripheral neuropathy and dementia frequently observed, as well as the visual impairment. For reasons which are not clear, however, the sequence of involvement of the pathways and the relative severity differ among patients, so that occasionally the organic mental syndrome or the peripheral neuropathy may be the predominating or even presenting features. In the spinal cord, lateral column involvement may appear early or late in the course, and may be masked by the peripheral neuropathy. The fact that this patient did not have pyramidal signs, therefore, is not an unusual phenomenon. It is noteworthy that the demyelination in the spinal cord is patchy,

so that levels showing sclerosis alternate with relatively normal ones.

An interesting aspect of the present case is the unusual sensory dissociation. The loss of vibration sense in the lower part of the body and the graphoanesthesia in the same areas contrast with the joint position sense (JPS) which was absent in the upper extremities, but only slightly reduced in the toes. These findings can hardly be explained by the traditional view which maintains that JPS and vibration are transmitted by the same system, viz. the dorsal columns. This view was based on the observation that JPS and vibration are lost together in three conditions (subacute combined, tabes dorsalis, and Friedreich's ataxia), in all of which the posterior columns are involved. However, it is now clear that the important similarity between these conditions is that they all affect the dorsal root ganglia, and the posterior column degeneration (as well, perhaps, as the peripheral neuropathy) is secondary to it.

Traditionally, vibration sense was thought to be a deep sensation. It now seems clear that this view was erroneous (7). Vibration excites cutaneous Pacinian corpuscles which have their own thickly myelinated direct lines to the spinal cord. These fibres are not involved solely with the transmission of vibratory stimuli. It has been stressed that a sense of vibration does not exist in the same meaning as other cutaneous sensations, such as temperature or touch, and it is probably true that vibration is the most unphysiologic of all sensations (7).

Upon reaching the spinal cord, signals that are triggered by vibration are transmitted along the posterior columns but are not limited to them. It is not clear which of the various pathways is responsible for the transmission of stimuli to consciousness, but it is unlikely that the dorsal funiculi are of primary importance. Based on careful clinical anatomic examinations, it was concluded that the posterior columns are not necessary for normal perception of vibration, and that the sensation can be lost with lesions of the spinal cord sparing the dorsal columns (7, 8).

Similar doubts relate to the role of the dorsal funiculi in JPS (8, 9). For this sensation too, dorsal columns are neither necessary nor sufficient. It is also clear that JPS and vibration can be affected selectively by anatomic lesions of the spinal cord, thus these two sensations cannot be presumed to be transmitted through the same pathways. It is thought that conscious proprioception is dependent upon the normal function of the dorsal spinocerebellar tract in the spinal cord, and particularly of

Morin's spinocervicothalamic pathway, which leads ipsilaterally to the ventrolateral nucleus of the thalamus (10).

Thus, it is clear that JPS and vibratory stimuli do not use the same pathways, and the dissociated loss of the two sensations encountered in this patient may be due to selective involvement of one of these. Other conditions affecting the spinal cord in a patchy or asymmetric manner which occasionally result in this dissociation include vascular (10), traumatic (9), and, perhaps most commonly, multiple sclerosis (11). That these modalities are frequently affected simultaneously is explained by the fact that they both make use of large-diameter, thickly myelinated nerve fibres, thus being exposed together to metabolic derangements.

How do we explain abnormal VEP in a patient with pernicious anemia and normal visual acuity? Could this dissociation represent damage to fibers which do not contribute to visual acuity, but which are still reflected in the VEP? Since originally reported by Halliday and his associates in 1972 (12), it has been demonstrated time and again that in many patients with multiple sclerosis who never have had any visual symptoms, the visual evoked potential is nevertheless abnormal, specifically it shows uni- or binocular delays, that is, the major positive deflection occurs later in these patients than in age-matched normal controls. The commonly accepted pathophysiologic explanation for these subclinical manifestations of multiple sclerosis is demyelination and plaque formation in the optic nerves. The neuropathology of experimental vitamin B₁₂ deficiency in monkeys has been studied by Agamanolis and his colleagues (13), who produced severe vitamin B₁₂ deficiency in rhesus monkeys. Two to three years after the institution of vitamin B₁₂ deficient diets, gross visual impairment appeared in five of the nine monkeys, while progressive spastic paralysis of the limbs developed later. The autopsy showed degeneration of the peripheral visual pathways in all, and of white matter of the spinal cord in four animals. Interestingly, in one animal, in whom B₁₂ later supplemented the diet, no abnormalities were found on postmortem examination, but in others the optic nerve sections showed marked loss of myelin and axons in discrete patches. At the globe, the lesion was near the temporal portion of the nerve, roughly corresponding to the papillomacular bundle, the lesion of which is thought to cause cecocentral scotoma in humans. Thus, it seems that demyelination associated with B₁₂ deficiency may be an explanation for VEP latency changes in this patient, similarly to many

multiple sclerosis patients (6) who have delayed VEP-s without visual symptoms. Probably VEP latency changes occur earlier than classical visual field defects when there is subtle optic nerve damage. Normal visual acuity and abnormal VEP latency is a very common finding of demyelinating disease.

In most patients reported in the literature, visual dysfunction, like the other manifestations of PA, improved as a result of specific therapy. In our patient, who had no symptomatic visual loss, the VEP latency improved as a result of therapy. In the only other report of a patient with PA who had delayed VEP (18), the effect of therapy was not mentioned. It would seem that such a short-term therapy as was instituted in our patient and in others reported in the literature (17), improvement in delayed conduction would be difficult to attribute to remyelination. Thus, if the improvement of VEP latency following a short course of therapy will be substantiated in other patients, one should consider alternate mechanisms of improvement such as direct role of vitamin B₁₂ in the optic nerve.

Acknowledgments

We thank M.B. Bender for suggesting VEP studies of this patient.

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Molluscum Contagiosum: Some New Histologic Observations

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Molluscum contagiosum, a disease caused by a large, brick-shaped virus ($300 \times 200 \times 100$ millimicrons) possessing a DNA-containing core (biconcave disc), is probably the easiest skin disease to diagnose microscopically because of the large pathognomonic cytoplasmic inclusion body which is readily identified, even by the novice. Perhaps because of this, until recently (1), detailed histologic studies and descriptions have been lacking in the medical literature. The current report represents microscopic findings following a detailed histologic analysis of these lesions.

Materials and Methods

Eighty-five skin biopsies from eighty-five patients showing the characteristic histopathology of molluscum contagiosum were reviewed from the files of The Skin and Mucous Membrane Biopsy Laboratory. All lesions had been fixed in Bouin's solution and stained with Hematoxylin and Eosin. Additional stains on selected cases included periodic acid-Schiff reaction (PAS) with and without diastase digestion, alcian blue (pH 4.5 and 0.5), Verhoeff's method for elastic tissue and the Feulgen reaction for deoxyribonucleic acid (DNA).

Results

Microscopically (Fig. 1), a typical, noninflamed and noninvoluting lesion was characterized by one or more cystic epidermal invaginations filled with a loose meshwork of keratin fragments and large, circular, homogeneous, predominantly basophilic bodies (measuring up to 35 microns in diameter), many of them showing a crescent-shaped, pyknotic

nuclear remnant at the margin. Many of these bodies were deeply eosinophilic at the base of these invaginations (Fig. 2). In some instances, eosinophilic bodies were also observed on and near the surface. The epidermis forming the sides of these invaginations was generally thinned and flattened, simulating the wall of a distended epidermal cyst. The subjacent epidermis was diffusely and markedly acanthotic assuming a bulbous configuration. The granular layer was thickened associated focally with coarse keratohyaline granules. Virtually all of the epidermal cells in this acanthotic portion were enlarged (Fig. 3). The keratinocytes of the basal layer and the lower half of the malpighian layer had abundant homogeneous to finely granular pale eosinophilic cytoplasm with large, ovoid to round vesicular nuclei, often having prominent nucleoli. As previously noted (1), the cytoplasmic and nuclear diameters of these cells were approximately 1.5 times greater than normal. Mitotic figures were identified only within the basal layer. As one progressed from the center of the malpighian layer to its junction with the granular layer, a relatively abrupt and striking change took place in the keratinocytes. Almost simultaneous cytoplasmic vacuolization and deeply eosinophilic condensation progressed after one or two layers to marked cytoplasmic enlargement and replacement by a distinct network of compartmentalized globules with clear centers and well-defined deeply eosinophilic rims ranging in size from less than one to four microns. Thus, the cytoplasm was virtually completely occupied by a large spherical molluscum body (up to 35 microns in diameter). The nuclei of these cells appeared as hyperchromatic crescent-shaped structures compressed at the periphery. The appearance of the infected keratinocytes in the granular layer was similar except for the presence of coarse keratohyaline granules compressed to the periphery, a more hyperchromatic nucleus and a gradual oblit-

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FIG. 1. Scanning power of lesion from palm demonstrating characteristic microscopic features as described. Obviously, the lesion has not developed at the site of a hair follicle (H & E $\times$ 40).

eration of structural markings within the molluscum body as one approached the surface of the granular layer, at which point it appeared as a diffusely homogeneous deeply eosinophilic body.

A well-defined, PAS-positive, diastase-resistant basement membrane zone was present about these acanthotic cystic invaginations. Adjacent to, outlining and paralleling the basement membrane zone, one always observed a conspicuous band of stromal proliferation of variable thickness consisting of loosely arranged thin collagen bundles and fibrocytes associated with a virtual absence of elastic tissue fibers. Vascular ectasia was often prominent within and about this zone of stromal proliferation. Increased amounts of glycogen were present in the basal layer and lower half of the malpighian layer. The eosinophilic molluscum bodies were weakly Feulgen-positive and the basophilic molluscum bodies were strongly Feulgen-positive. The basophilic molluscum bodies located within the superficial layers of the cystic invaginations stained with alcian blue at pH 4.5. It was also noted that no

melanin granules were present within the keratinocytes of the involved acanthotic epidermis in both Caucasian and Negro skin.

Other interesting findings were infection of the epidermis overlying an intradermal nevus (Fig. 4) and infection of a relatively deep epidermal cyst involving both reticular cutis and subcutaneous tissue by the molluscum contagiosum virus (Fig. 5). Of added significance was the presence of a characteristic zone of stromal proliferation as described in typical molluscum contagiosum lesions, limited only to the portion of the wall showing the infection. This lesion clinically was a firm subepidermal nodule of several weeks' duration located on the anterior neck just beneath the skin and to the right of the midline in a 7-year-old Caucasian boy. The overlying skin surface was normal and there had been no recent changes prior to removal. The clinical impression was a sebaceous cyst. The patient also had several characteristic molluscum contagiosum lesions in the same general area.

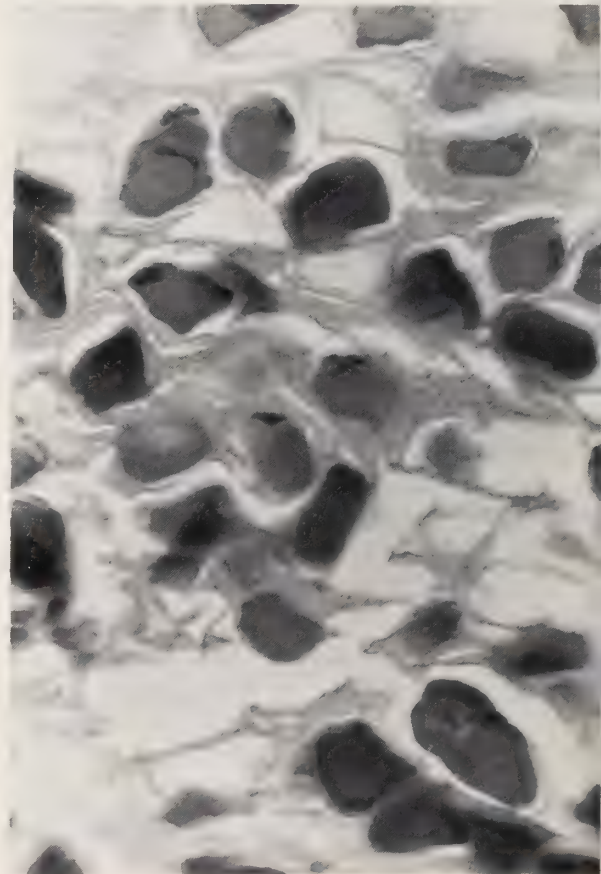


FIG. 2. High magnification of base of invagination showing mixture of large cytoplasmic eosinophilic (lighter-staining) and basophilic (darker-staining) molluscum bodies with eccentric pyknotic nuclear fragments interspersed among keratin meshwork (H & E $\times$ 400).

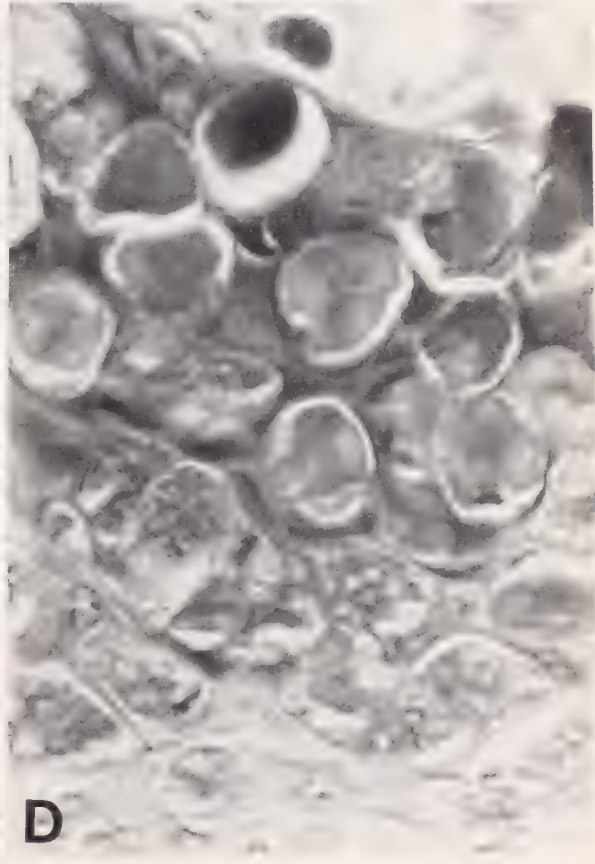
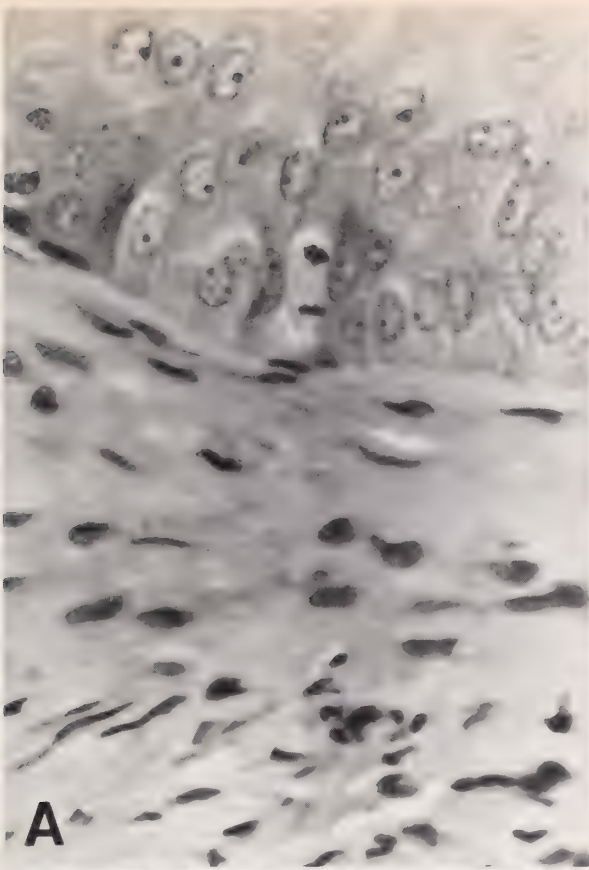


FIG. 3. High magnification of infected epidermis detailing progressive changes. Please note stromal proliferation, enlarged keratinocytes with vesicular nuclei, pale cytoplasm and mitotic activity in basal layer (A), cytoplasmic enlargement, vacuolization, condensation and compartmentalization in center of malpighian layer (B), gradual obliteration of structural markings in molluscum body at junction of malpighian and granular layers (C) and characteristic, homogeneous, eosinophilic cytoplasmic inclusion bodies with eccentric, hyperchromatic nuclei and keratohyaline granules in granular layer (D) (H & E $\times 400$).

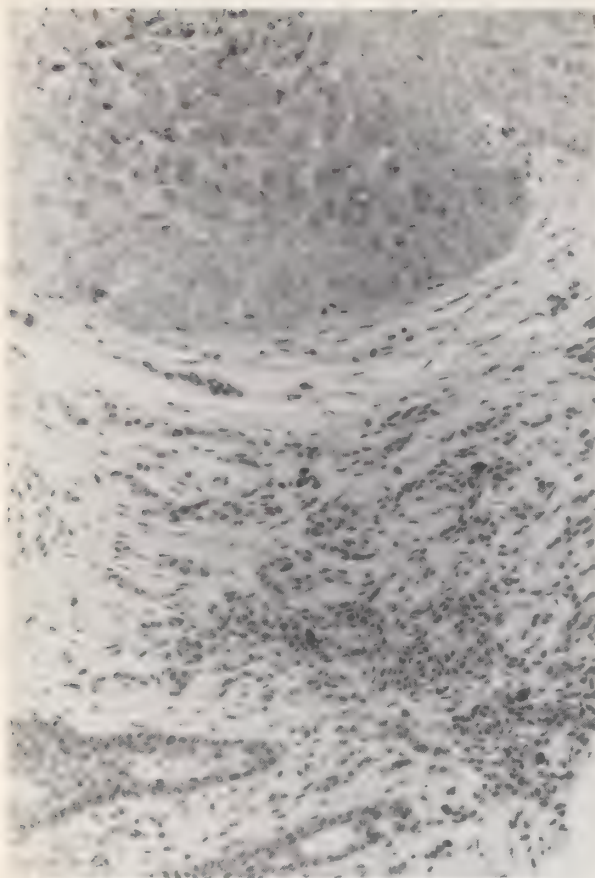


FIG. 4. Molluscum contagiosum infection of epidermis overlying an intradermal nevus (H & E $\times 100$).

Discussion

The basic light microscopic features of molluscum contagiosum are well described (2) and its ultrastructure has been studied in detail (3). The mechanism of infection is still controversial (1). Evidence indicates that infection occurs externally and that the basal cells of the epidermis are initially involved. My findings support this concept but differ in several respects from those of others (1). I saw no evidence of intranuclear inclusion bodies within malpighian keratinocytes. No eosinophilic cytoplasmic particles resembling trichohyaline were seen within the granular layer. No features suggesting follicular neogenesis were noted. For the most part, nonadnexal epidermal keratinocytes were primarily involved, although epidermal trichilemmal keratinocytes were also infected in a number of instances. It does not seem to have been previously recognized that the entire keratinocyte with its molluscum body and pyknotic crescent-shaped nucleus is desquamated into the cystic invagination, thus explaining the absence of parakeratosis. Since

visibly infected cells can not keratinize (3), the abundant amount of keratin which is present within these cystic invaginations either must have been produced by apparently uninfected keratinocytes and/or infected keratinocytes before visible evidence of infection and interference with keratinization.

The presence of increased amounts of epidermal glycogen was expected. This is a nonspecific finding observed in a variety of specific and nonspecific inflammatory dermatoses, often associated with epidermal hyperplasia, as well as in benign and malignant neoplasms of keratinocytes. This is probably a reflection of altered cell metabolism. As mentioned by others (2), the degree of Feulgen positivity is most likely directly related to the degree of destruction of the protein shell or capsid surrounding the viral nucleic acid core. Contrary to the findings of others (2), the molluscum bodies did not become basophilic until shortly after desquamation into the cystic invaginations. Since maximum Feulgen positivity was observed with maximum basophilia of the molluscum bodies, the degree of basophilia was also probably directly dependent upon the degree of capsid destruction. It was interesting that the basophilic molluscum bodies within the superficial layers of the cystic invaginations stained with alcian blue at pH 4.5. This is probably indicative of further destruction of the capsid, allowing staining of the nucleic acid core (nucleoid).

Further evidence of altered metabolism was shown by the absence of melanin granules within Negro as well as Caucasian keratinocytes involved by molluscum contagiosum. Similar findings have been noted in the clear cell acanthoma.

Epithelial-stromal interaction is well known (4). Classic examples are the zone of stromal proliferation about the periphery of basalioma cells in basal cell carcinomas and about the normal pilary complex. Stromal proliferation is a definite and conspicuous component of all noninflamed and noninvoluting molluscum contagiosum lesions which, until recently (1), has not been generally recognized. Contrary to the beliefs of these authors (1), I do not believe that it has any histogenetic relationship with follicular neogenesis. Although it is quite possible that the combination of rapid epidermal hyperplasia and cell hypertrophy produced sufficient physical tension in the papillary cutis to cause reactive stromal proliferation (4), this seems unlikely since similar stromal changes are observed about the normal pilary complex as well as basal cell carcinomas. The common denominator of all these conditions, normal, neoplastic or inflamma-

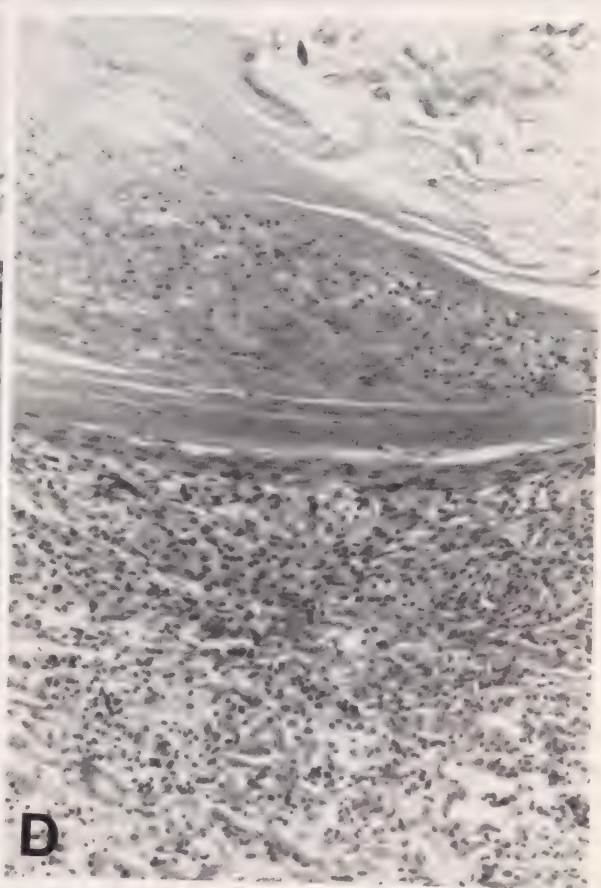
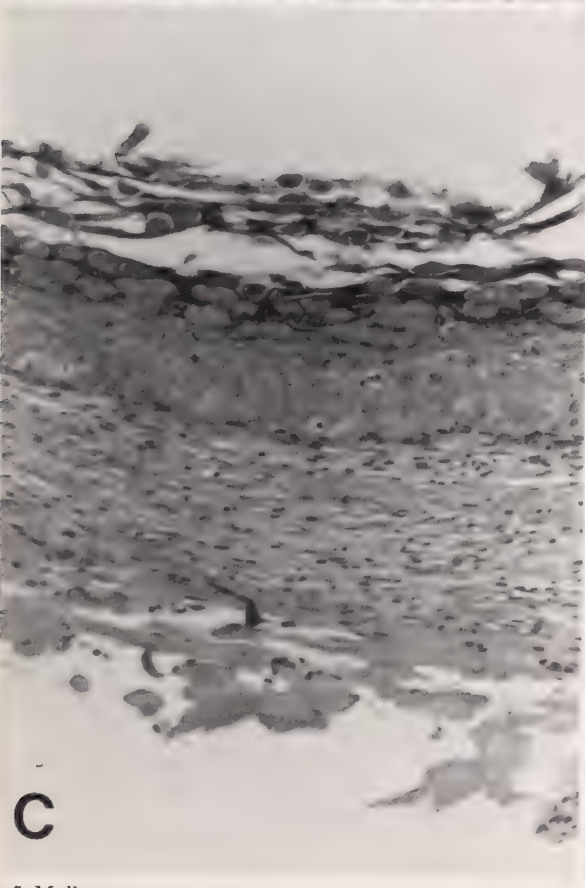
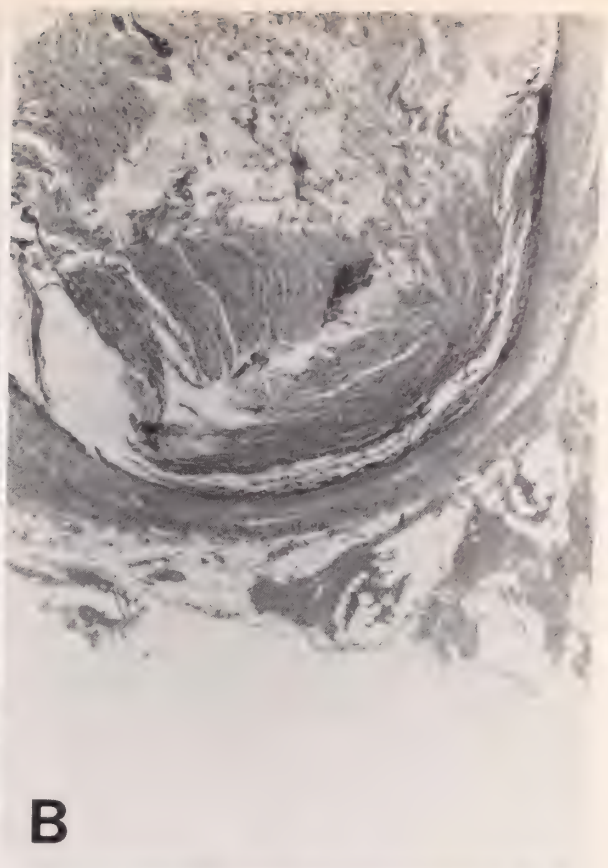
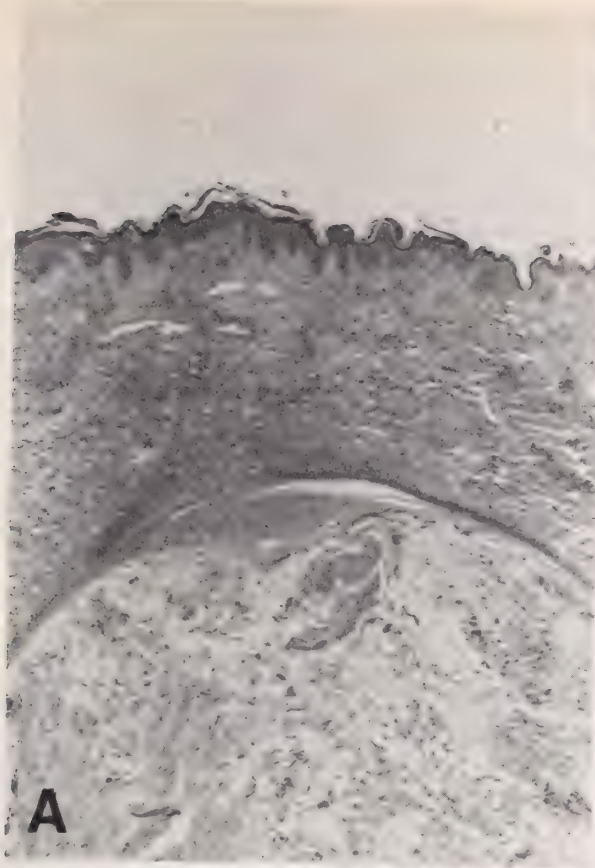


FIG. 5. Molluscum contagiosum infection of epidermal cyst showing upper (A) and lower (B) halves (H & E $\times 20$), characteristic changes of wall with stromal proliferation (C, H & E $\times 100$) and absence of stromal proliferation about inflamed wall without evidence of molluscum bodies (D, H & E $\times 100$).

tory, appears to be the basal or peripheral keratinocyte. This suggests that this cell, through some unknown mechanism, has the ability to induce collagen biosynthesis. Although buds of hair follicles were identified in association with a number of these lesions, they were interpreted as residual portions of preexisting infected hair follicles, and not evidence for follicular neogenesis (1).

Although infection of an epidermal cyst has been previously reported (5), its authenticity has been refuted (6). For similar reasons, the authenticity of a more recent report (7) remains doubtful. In my case, however, there is absolutely no room for doubt about its authenticity. In fact, it is surprising that infections of other dermatoses with this common virus have not been reported. Infection of the epidermis overlying an intradermal nevus, although apparently not previously reported, is probably not uncommon. This phenomenon is relatively common with warts (8).

Summary

Histologic analysis of 85 cases of molluscum contagiosum has been detailed. No evidence for follicular neogenesis is noted. No intranuclear inclusion bodies were observed. Infection seems to start in the basal layer with progressive outward orderly maturation and intracavitary desquamation of keratin and molluscum bodies with attached pyknotic nuclear remnants. The degree of Feulgen positivity and basophilia of the molluscum bodies appears to be directly related to the degree of destruction of the capsid. Basophilic and eosinophilic molluscum

bodies were observed within the cavity. The superficially located basophilic bodies stain with Alcian Blue at pH 4.5 which may be indicative of further destruction of the capsid. The infected keratinocytes contain increased amounts of glycogen and do not seem to be able to ingest melanin. Stromal proliferation is a conspicuous feature adjacent to a well-defined PAS-positive, diastase-resistant basement membrane zone in all noninflamed and non-involuting lesions. Current evidence suggests that the basal or peripheral keratinocyte, through some unknown mechanism, might have the ability to induce collagen biosynthesis. Authentic documentation of infection of an epidermal cyst and the epidermis overlying an intradermal nevus has been presented.

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Respiratory Distress, Pericarditis and Inappropriate Antidiuretic Hormone Secretion in a Patient with Infectious Mononucleosis and Gaucher's Disease

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Although infectious mononucleosis is frequently associated with mild respiratory symptoms, the pulmonary manifestations rarely dominate the clinical picture. We present a patient with the adult form of Gaucher's disease who developed acute pneumonitis with respiratory distress associated with infectious mononucleosis. During the illness, the patient also manifested pericarditis and the syndrome of inappropriate antidiuretic hormone (ADH) secretion. The latter has not previously been reported to occur in Gaucher's disease or in infectious mononucleosis. The possible interaction of infectious mononucleosis and Gaucher's disease is explored.

Case Report

A 20-year-old white male with Gaucher's disease (diagnosed at age 3, splenectomy at age 11 and a history of mild asthma) was asymptomatic until four days prior to admission to The Mount Sinai Hospital, New York, when he developed fever and myalgias. On the day of admission, he complained of severe low back pain, chest tightness and shortness of breath. Temperature was 102.4° F and respiratory rate was 24 per minute. There were shoddy posterior cervical and axillary lymph nodes. No wheezes or rales were audible. The liver was palpable 15 cm below the costal margin. Admission chest roentgenogram was normal. White blood count was 21,000, of which 18% were lymphocytes. The patient was treated with ampicillin, oxacillin, and heparin. By the fourth day, the respiratory distress had increased and the patient was transferred to the Respiratory Intensive Care Unit. Ar-

terial blood gases while breathing room air were PO₂ 55 mm Hg, PCO₂ 37 mm Hg, and pH 7.45. White blood count was 22,900 with 35% lymphocyte of which 11% were atypical. Lung scan showed no evidence of pulmonary emboli and heparin was discontinued. On the sixth hospital day, the patient developed diffuse nonpruritic erythematous rash and the oxacillin and ampicillin were discontinued. Chest roentgenogram showed bilateral small pleural effusions, diffused interstitial infiltrations, and an increase in the size of the cardiac silhouette. A pericardial friction rub was heard and an echocardiogram confirmed the presence of a small pericardial effusion. The mono spot test, which was negative on admission, became positive on the 13th hospital day. Heterophil was positive 1:448 and 1:112 after absorption with guinea pig kidney. EB virus titer, drawn on the third hospital day, was 1:100. Viral and PPLO acute and convalescent titers were negative. On admission, the serum sodium was 132 meq/L and decreased to 123 meq/L by the 8th hospital day. The serum osmolality was 261, urine osmolality 413 and the urine sodium was 42 meq/L. Serum cortisol levels were normal. With only supportive therapy, by the 30th hospital day the patient became afebrile, the shortness of breath abated, the pericardial friction rub was no longer audible, the arterial blood gases returned to normal, and serum sodium and osmolality were normal.

Pulmonary function tests were performed on the 6th, 16th and 35th hospital day and six weeks and seven months after discharge and are presented in Table I. The initial study revealed a restrictive pattern of pulmonary dysfunction with marked reduction in the forced vital capacity and single breath diffusing capacity to 50% and 43% of pre-

TABLE I
Pulmonary Function Data

	FVC (liters)	% Predicted	FEV ₁ l/sec	FEV ₁ /FVC	D _L CO _{SB} (% Predicted)
Day 6	2.98	50	2.29	.76	43
Day 16	4.84	81	3.70	.76	54
Day 35	5.40	91	3.60	.67	52
6 wks after discharge	6.10	103	3.70*	.61	59
7 mos after discharge	6.27	106	3.77*	.60	74

* Following bronchodilators there was a 13% increase in FEV₁.

dicted, respectively. FEV₁/FVC was normal. Over the ensuing eight months there was gradual improvement in the FVC and D_LCO, but the D_LCO has remained below normal. Since pulmonary function tests had not been performed prior to his illness the decreased FEV₁/FVC may reflect residual airways dysfunction or be consistent with his history of asthma.

Discussion

Mild respiratory complaints are common in patients with infectious mononucleosis. The admitting diagnosis of 19 of 300 patients with infectious mononucleosis, reported by Reed, was "atypical pneumonia" (1). However, respiratory distress was not described in any of these patients. Radiographically, infectious mononucleosis has been associated with patchy pneumonitis, pleural effusion, and hilar lymph node enlargement (2, 3). In the few patients with infectious mononucleosis from whom lung tissue was available for pathologic study, local or diffuse interstitial infiltration by mononuclear cells or atypical lymphocytes was seen (4). Recently, there have been several reports of pulmonary involvement in Gaucher's disease. Infiltration of the alveolar interstitium, as well as alveolar filling with Gaucher cells, has been noted in the adult form of Gaucher's disease. Such involvement may cause subclinical pulmonary dysfunction or be massive leading to death or chronic pulmonary disability (5, 6). It is a matter of conjecture whether the patient reported here was rendered more susceptible to the unusual pneumonitis and respiratory distress of infectious mononucleosis because of subclinical pulmonary infiltration by Gaucher cells. This supposition may be supported by the persisting pulmonary function abnormalities more than seven months after recovery from infectious mononucleosis. Further, pulmonary function testing of asymp-

tomatic patients with Gaucher's disease and patients with infectious mononucleosis is necessary to delineate the prevalence of respiratory involvement in these diseases.

Pericarditis has been described in both infectious mononucleosis (7, 8) and Gaucher's disease (9). Approximately 20 patients with infectious mononucleosis and pericarditis have been reported. In one patient, pericarditis was the sole manifestation of the disease (8). Roberts and Fredericks reported a 30-year-old male with Gaucher's disease who developed severe pulmonary hypertension from obstruction of the pulmonary capillaries by Gaucher cells (9). That patient had multiple episodes of acute pericarditis over a 13-month period and later died of hemopericardium and cardiac tamponade. In that report, no etiology could be definitively assigned since the patient did not have Gaucher cells in the pericardium. The more frequent association with infectious mononucleosis and the absence of recurrence suggests that in our patient the pericarditis was a manifestation of infectious mononucleosis.

A final unusual aspect of the patient's illness was the presence of the syndrome of inappropriate ADH secretion. The patient fulfilled the criteria for the syndrome manifesting hyponatremic and hypoosmolar serum while his urine was not maximally dilute and the urine sodium was greater than 30 meq/L. The patient had no edema and there was no evidence of congestive heart failure, renal or adrenal disease. His hyponatremia corrected spontaneously as his respiratory status improved. There has been no previous report of the syndrome of inappropriate ADH in Gaucher's disease or in infectious mononucleosis. Many pulmonary processes including neoplasms, tuberculosis, fungal infections, bacterial and viral pneumonias have been associated with the syndrome of inappropriate ADH secretion (10). The pneumonitis associated with infectious mononucleosis should now be added to this list.

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Epidural Blood Patch for Spinal Headache

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Introduction

Headache is a common complication of a spinal dural puncture. It occurs following spinal anesthesia, inadvertent dural puncture during attempted epidural anesthesia, and after diagnostic myelography. The headache may be severe and incapacitating. In addition, other neurologic symptoms—diplopia, tinnitus and nausea—may develop (1). Subdural hematomas have also been reported (2).

Many therapeutic modalities have been recommended: complete bed rest, forced fluids, abdominal binder, epidural injections of saline, and analgesics (3). Some do alleviate the headache, but the relief is often temporary and incomplete. Epidural blood patch has been found to be very effective and is becoming a popular treatment of spinal headache (4). The following is our experience with this treatment in eight patients. Three are reported in detail because of special clinical features.

Materials and Methods

Eight female patients, ages 18 to 35, were referred to us for treatment of spinal headache. Four had had spinal anesthesia, three inadvertent puncture of the dura during attempted epidural anesthesia, and one a myelogram for low back pain. All were bedridden by headache, and often by dizziness and nausea, for at least three days. Two patients developed diplopia and tinnitus. Complete bed rest, forced fluids, abdominal binder, and codeine preparations afforded only partial or temporary relief. Epidural injection of blood was then offered and all patients consented, with the exception of the fifth patient (Table I).

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With an I.V. of Ringer's lactate running, each patient was turned on her side and the intervertebral space previously entered was identified. To minimize chances of infection, the epidural space above or below this space was entered. Meanwhile, an assistant prepared the patient's antecubital fossa and 10 ml of blood were drawn from a vein, using sterile precautions and injected into the epidural space. The needle was then withdrawn and the patient turned flat on her back. One litre of Ringer's lactate was administered I.V. within the next 30 minutes. The patient was then asked to get up, walk and evaluate the treatment. All patients were given cephalosporin 2 gms a day prophylactically for three days.

Results

Six patients experienced immediate and permanent relief. One experienced recurrent headache a few hours later, but a second epidural blood injection resulted in a complete and permanent relief (Patient No. 2). One patient had no relief (Patient No. 8).

The following are three Case Reports with special clinical features:

Case Report

Patient No. 2

An 18-year-old prima gravida was scheduled for an emergency cesarean section for cephalopelvic disproportion. During attempted epidural anesthesia with a 16 gauge Touhy needle, the dura was inadvertently punctured. The procedure was abandoned and general anesthesia was administered uneventfully. Twenty-four hours later the patient developed severe headache and was completely incapacitated and bedridden. Conservative management for three days was to no avail. She then

TABLE I
Results of Epidural Blood Patch in Eight Patients.

Patient No.	Age	Headache Cause	Needle Size	Results
1	25	inadvertent spinal tap	16	immediate and permanent relief
2*	18	inadvertent spinal tap	16	immediate relief recurrence of headache
3	32	myelogram	?	immediate and permanent relief
4	29	spinal anesthesia	22	immediate and permanent relief
5*	25	spinal anesthesia	22	bloody epidural tap resulted in immediate and permanent relief
6	30	inadvertent spinal tap	17	immediate and permanent relief
7	19	spinal anesthesia	22	immediate and permanent relief
8*	35	spinal anesthesia?	22	no relief. history of hypertension and chronic headache

* See Case Report

consented to and was given epidural blood injection. Thirty minutes later she was able to get up and walk around free of symptoms except for mild dizziness. The next morning, at a follow-up visit, she was again found in distress complaining of severe headache as before. Careful questioning revealed that five hours after the epidural blood injection the patient went to the bathroom to move her bowels. Because she was constipated, she strained and shortly thereafter the headache recurred. The patient consented to a second epidural blood injection and this time the relief was immediate and permanent. A laxative was prescribed and the patient was discharged two days later free of symptoms.

Patient No. 5

A 25-year-old nurse had spinal anesthesia with a 22 gauge needle for low forceps delivery. The next morning, after a short walk, she developed severe headache relieved only by lying down. In spite of conservative management for three days, the headache persisted. The patient refused epidural blood injection but agreed to epidural saline injections. When the epidural space was entered, blood appeared at the needle's hub. The needle was withdrawn slowly until the blood flow ceased and then an epidural catheter was inserted. Saline was not injected. The patient was asked to remain in bed for about 30 minutes and then get up and walk. The headache was completely gone and she was asymptomatic for the rest of her hospital stay.

Patient No. 8

A 35-year-old full term multipara was admitted to the hospital because of a blood pressure of 160/90 and proteinuria. She had had a long history of hypertension and had been treated with low salt diet. Labor was successfully induced with oxytocin, and spinal anesthesia with a 22 gauge needle was given to facilitate low forceps delivery. During her stay in the hospital she complained of mild headache which was presumed to be due to the spinal tap. She was treated conservatively and discharged after three days. A week later she called her obstetrician and complained of a "nagging" headache. She was invited back to the hospital and offered an epidural blood injection. She agreed and the injection was given in the usual manner. Following the injection the patient denied any beneficial effect. Upon careful questioning, she admitted that she had been suffering from chronic headache and that it had been diffuse in character and not related to posture.

Discussion

The underlying cause of spinal headache is thought to be low cerebrospinal fluid (CSF) pressure caused by seepage of CSF through a dural rent (5). The incidence of spinal headache is related to the gauge of needle used. The incidence with 25, 24 and 18 gauge needles was reported to be 2.2% (6), 8% (7), and 24% (8), respectively. Following a myelogram, it was 44% (9) and after inadvertent punc-

ture of the dura during attempted epidural anesthesia 76.9% (10).

Spinal headache is usually self limited. Most patients recover within a week (24% within two days, 29% between two to four days, and 19% between five to seven days) (11). Mean duration is four days (3). However, occasionally spinal headache may linger for months and completely incapacitate the patient.

Our report of Patient No. 8 underscores the importance of cautious diagnosis of spinal headache. Headache is not an uncommon symptom following surgery, but only a spinal headache responds to epidural blood injection. This headache, characteristically, is occipitofrontal and gets worse in the erect posture (1). It usually appears 24 hours after the dural puncture (4) and may be accompanied by one or more of the following: nausea (22%), blurred vision (10%), tinnitus (5%) and stiff neck (5%).

A wide array of therapeutic measures have been recommended (3): analgetics, analeptics, antihistamines, narcotics, sedatives, I.V. fluids, anesthetics, I.V. ethanol, abdominal binder, and epidural or subarachnoid injection of air, saline, glucose, or steroids. These measures usually fail or afford temporary or partial relief only. In contrast, epidural blood injection is completely effective within 24 hours in nearly 100% of the patients (4, 12).

When compared to epidural saline injection, epidural blood injection has been proven to be superior: spinal headache, following dural puncture with a 25 gauge needle, was cured after saline or blood injections in 60% and 100% of the patients, respectively. When the headache followed a dural puncture with a 17 gauge needle, saline injections failed to cure any patients while blood injections cured permanently 73% of the patients (13). Epidural saline injections apparently reduce CSF seepage through the dural rent by increasing epidural pressure. However, the pressure returns to base line three to ten minutes after the injection (14). This explains the temporary relief after saline injection.

The recurrence of the headache after the first epidural blood injection in Patient No. 2 could be caused by the fragility of the gelatinous layer formed by the blood clot over the dural rent. Animal studies showed no cellular reaction after epidural blood injection until 48 hours later (15). It is conceivable, therefore, that straining during the first two days after the treatment may break the seal over the dural rent and cause a recurrence of the headache. Patients should be instructed to avoid straining and take a laxative for at least 48 hours following treatment.

Concern has been expressed as to potential dangers of epidural blood injection (16). Human and animal studies do not support it. Although backache (1, 4), radicular pain (17, 18), dizziness (12), parasethias, facial palsy, and fever (1) have been reported to follow epidural blood injection, these complications are mild and respond to conservative measures. No cases of arachnoiditis occurred in a large series (15).

Successful epidural anesthesia has been reported after epidural blood injection (1, 19). One report describes a case of a spinal headache, following inadvertent puncture of the dura, that required two epidural blood injections before it was cured. One week later epidural anesthesia was given for an emergency dilation and curettement resulting in a normal onset and level (19). Another author reports successful epidural, spinal and caudal blocks 105, 139, and 380 days, following epidural blood injection, respectively (1). Animal studies support these observations: epidurally injected blood was completely absorbed within two weeks and local histologic changes did not differ from those seen after a simple dural puncture (15).

In conclusion, our experience with epidural blood injection confirms that it is indeed an effective therapeutic modality for spinal headache. However, the diagnosis of spinal headache should be cautiously made, and patients should be instructed to avoid straining and to take a laxative for at least 48 hours following the treatment.

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Primary Tuberculosis of the Ileum Roentgen Features

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Abstract

Intestinal tuberculosis in the absence of active pulmonary disease is a rare entity in nonendemic areas. The roentgen features are indistinguishable from the findings in regional enteritis (Crohn's Disease) and/or carcinoma, metastatic or primary. However, in clinically atypical cases, the diagnosis should still be entertained particularly in lesions of the distal ileum and ileocaecal region. A case of primary ileal tuberculosis is presented to illustrate these features.

Case Report

A sixty-two-year old white female was admitted to The Mount Sinai Hospital with a two-week history of right lower extremity pain and swelling. She had been admitted five months previously complaining of pleuritic chest pain, shortness of breath, and crampy abdominal pain of many months duration. Chest x-ray at that time showed a left lower lobe infiltrate and, since ventilation perfusion scan of the lungs was suggestive of pulmonary embolism, she was anticoagulated. Three weeks later, a cavity developed in the left lower lobe which was believed to be secondary to the infarction. Treatment with oral penicillin resulted in resolution of the pulmonary disease. In an effort to localize the abdominal pain, barium enema, upper gastrointestinal series and oral cholecystography were performed. All studies were negative.

The past medical history elicited on the present admission revealed no travel abroad. She was born in the United States. On physical examination, the patient was pale and significant weight loss was noted since her previous admission. An abdominal mass was not palpated. The stool was guaiac negative. Smear and culture of induced sputum failed to reveal evidence of acid fast bacilli. The chest x-ray was unremarkable. A repeat upper gastrointestinal

series with small bowel study revealed partial obstruction in the mid ileum due to a short stenotic segment (Figs. 1, 2). At laparotomy, an infiltrative lesion was found two inches proximal to the ileocecal valve. The serosa was studded with hard waxy appearing nodules. About twenty inches proximal to the ileocecal valve, a second similar appearing lesion was found which invaded the mesentery. There were multiple hard nodules in the mesentery. Proximal to these lesions, the small bowel was dilated. The diseased segments of small bowel were resected in continuity with the cecum and a side-to-side anastomosis between the ileum and the right colon was performed.

On pathologic examination, the mucosa was found to be quite rough and nodular but without ulceration. The mesenteric fat was edematous. Two small white fleshy lymph nodes were found. The external surface of the small bowel was granular. There were segmental areas of transmural necrotizing granulomatous inflammation containing acid fast bacilli. There was focal stenosis with proximal dilatation. The adjacent fat and numerous lymph nodes showed fibrosis with necrotizing granulomata and acid fast bacilli. The segment of colon and appendix showed no significant change. Subsequently, cultures from the operative specimen grew mycobacterium tuberculosis.

Discussion

The incidence of pulmonary tuberculosis in the presence of intestinal tuberculosis is unclear. In several studies, consistently less than 50% of pa-

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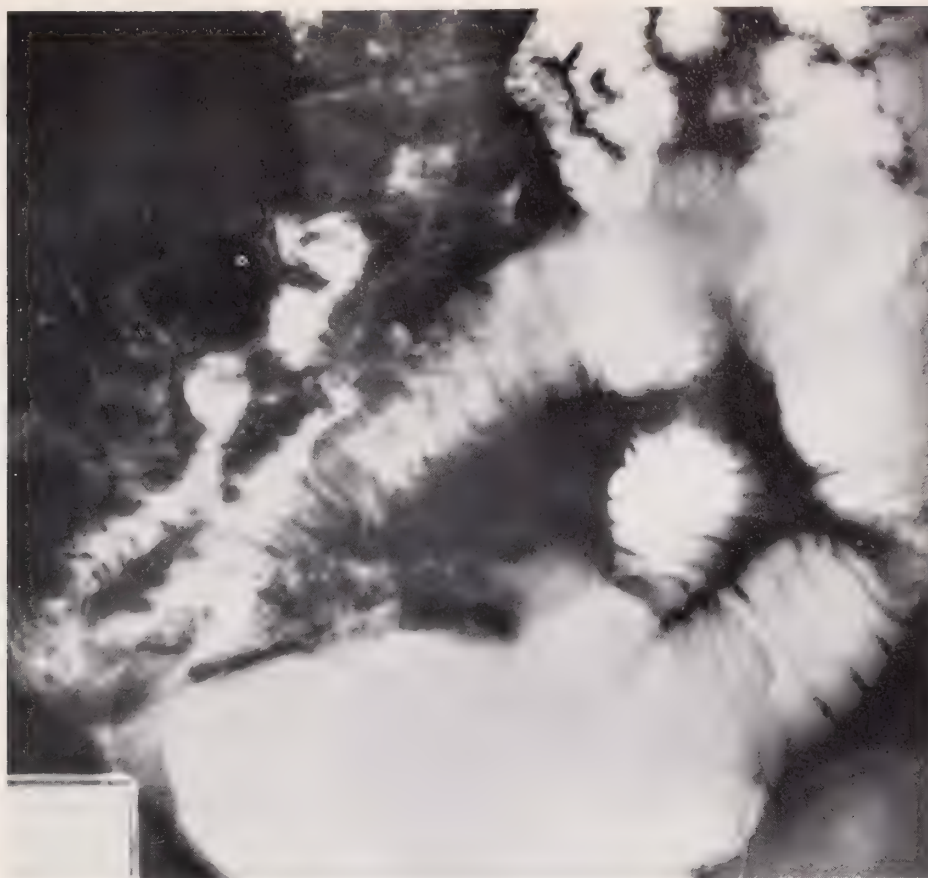


FIG. 1. Jejunal loops in the left upper abdomen are completely normal. The ileum, however, is dilated down to the right lower quadrant where an abrupt transition to collapsed small bowel is evident. A five centimeter segment of marked narrowing is evident within the mid-ileum. This portion of the bowel is fixed. The contours are scalloped because of nodular defects in the wall. At the proximal margin of the lesion the barium is hazy as if diluted by secretions but no discrete ulcerations are identified.

tients proven to have intestinal tuberculosis had radiographic evidence of active pulmonary disease (1). Bovine *Mycobacterium Tuberculosis* is virtually nonexistent in the United States although it still accounts for much intestinal disease in countries where milk is not pasteurized.

Tuberculous infection of the gastrointestinal tract is distributed according to the richness of gastrointestinal lymphoid tissue. Thus, the most commonly affected area (80%–90% of cases) is the ileocecal region where the highest density of Peyer's patches is found. The bacilli are absorbed by intestinal mucosal cells and proliferate in the submucosal lymphoid tissue. Typical tubercles of epithelioid cells and lymphocytes are formed in the submucosa following the distribution of lymphatic channels. These tubercles may expand toward the bowel lumen resulting in mucosal ulceration, usually manifested as circumferential or transverse ulcers caused by the transverse orientation of the submucosal lymphatics. Rarely, these ulcers may

be longitudinal and short if there is ulceration of tubercles within the Peyer's patches. This may be followed by a transmural granulomatous process which thickens the bowel wall and leads to fibrosis and stricture. Regional lymph nodes become involved when submucosal tubercles undergo necrosis and caseation with release of organisms into intramural lymphatics.

Three gross pathologic forms of intestinal tuberculosis are described. The ulcerative form is considered the most common. The hypertrophic form is more common in the ileocecal area and colon than in the mesenteric small bowel. This particular form is frequently associated with a negative chest x-ray and is considered by many to be a primary tuberculous infection (2). The bowel wall, the mesentery and the surrounding lymph nodes are thickened and matted together often presenting as a palpable abdominal mass. The bowel lumen may be stenotic. Hyperplastic nodular mucosa and a large intraluminal inflammatory mass resembling a neoplasm



FIG. 2. A close-up of the diseased ileal segment clearly demonstrates the nodular defects seen both in profile and en face. These represent areas of submucosal granulomatous inflammation which, with reactive fibrosis, produce marked narrowing of the lumen.

may occur. We believe that this case falls into this category and represents a primary tuberculous infection of the ileum. Combined features of both forms of tuberculosis enterocolitis may occur in any area and lead to an ulcerohypertrophic form.

Roentgen Features

The earliest recognizable features of the disease involve inflammation of the small bowel submucosa which leads to stiffening and thickening of the folds. The folds may appear nodular. The mucosa can be diffusely ulcerated. When visualized, discrete ulcers are often stellate with convergence of mucosal folds (1). In more extensive disease, large transverse ulcers can be demonstrated extending into the submucosa. Enteroenteric fistulae and enterocutaneous sinuses may develop in advanced disease. Hypertrophy is characterized by mucosal nodularity, mass formation, rigidity and fixation of the bowel, irregularity of the bowel contour and separation of bowel loops.

Distortion of the ileocecal region consisting of retraction and shortening of the cecum, localized stenosis of the cecum at the level of the ileocecal valve, and the appearance of an inflammatory mass in the cecum is a frequent finding (2). Changes in the ileocecal valve include thickening of the valve lips by granuloma formation and wide gaping of the valve itself associated with narrowing of the immediately adjacent ileum (Fleischner's sign) (3, 4). Narrowing of the terminal ileum may be secondary to rapid emptying of the involved segment whether filled in a retrograde or antegrade fashion and represents irritability and spasm in the acute inflammatory phase of the disease. This appearance is known as Steirlin's Sign and is not specific for tuberculosis (5). A similar appearance may be the result of stricture in advanced ileocecal tuberculosis. A case seen in this hospital demonstrates many of the changes of moderately advanced ileocecal tuberculosis (Fig. 3).

There are no radiographically specific diagnostic

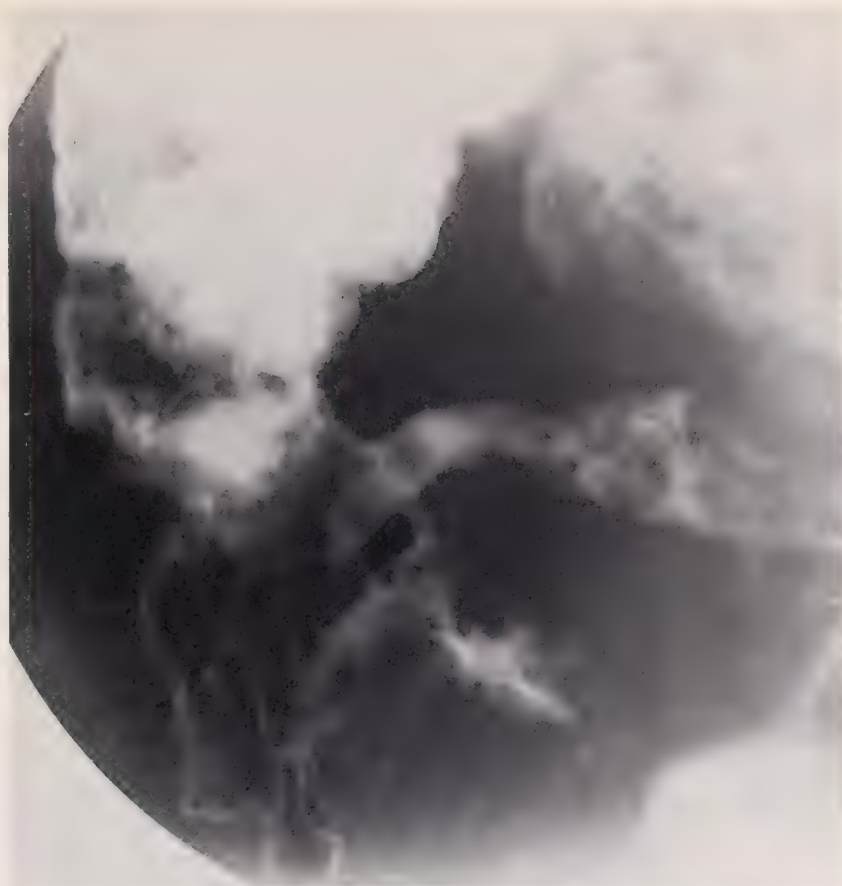


FIG. 3. The involved distal ileum is separated from the remainder of the small bowel by a mass in the mesentery. The terminal ileum is narrowed, and the valvulae conniventes are effaced. No discrete ulcers are visualized. The contour is smooth. The cecum is narrowed. The caput coli is compressed by an inflammatory mass in the mesentery. The lips of the ileocecal valve are not visualized.

features available for definitive preoperative diagnosis of intestinal tuberculosis. In the ileocecal region, the appearance is virtually identical to Crohn's disease. In the mesenteric small bowel differentiation from infiltrating neoplasm is almost impossible especially in the hypertrophic type such as the case presented. However, it is important to, at least, consider the possibility of tuberculosis in atypical presentations of distal small bowel lesions, since this is the only disease in the differential which is curable by medical therapy.

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Bouin's Solution Versus Formalin Fixation in Cutaneous Histopathology

JOHN KWITTKEN, M.D.

It has been stated (1, 2) and generally accepted that ten percent neutral buffered formalin is the best overall fixative for tissue pathology and that Bouin's solution has deleterious effects on tissue with the passage of time caused by residual picric acid. I have been practicing pathology for more than two decades and have had almost equal experience with both fixatives. During the first half of my career, I used formalin exclusively and I have been using only Bouin's solution for the past 11 years in my laboratory. Contrary to what has been generally accepted, fixation in Bouin's solution is far superior to formalin fixation. In order to obtain an objective evaluation, a study was performed which represents the subject of this report.

Materials and Methods

One hundred consecutive skin biopsies that were large enough to section in half were performed in my dermatology office. One half of each biopsy was placed immediately in Bouin's solution (picric acid, saturated aqueous solution, 75.0 ml; formaldehyde, 37%-40%, 25.0 ml; glacial acetic acid, 5.0 ml) and ten percent neutral buffered formalin. Fixation time before processing ranged from one hour to eight hours. The specimens were processed in the Auto-technicon Ultra and sectioned with the microtome. Small specimens were processed on the short cycle (four hours) and large specimens were processed on the over-night cycle (sixteen hours). Stains utilized included Hematoxylin and Eosin, Verhoeff's elastic-Van Gieson, Periodic Acid-Schiff (with and without diastase digestion), Alcian Blue (pH 4.5 and 0.5), Fite-Faraco, Perl (for iron), Polychrome Methylene Blue (for mast cells), Methyl Violet (for amyloid)

and the Von Kossa (for calcium). All stained slides were examined histologically at periodic intervals ranging from one hour to twelve months from the time of preparation. In addition, all paraffin blocks were resectioned at a period ranging from six to twelve months from the time of the initial sectioning, stained in the same fashion as the original sections, and examined within a 24-hour period.

Results

Except for excellent preservation of red blood cells with formalin fixation and hemolysis with Bouin's solution in many instances (Fig. 1), Bouin's fixation proved to be superior in several ways. The most important advantage was preservation of nuclear detail (Figs. 2-4). With formalin fixation, nuclei appeared smudged with loss of definition of the chromatin network and nuclear membrane. Many nuclei appeared pyknotic with linear distortions. The latter was especially true of the cells within the basal layer of the epidermis. With Bouin's fixation, the chromatin network and nuclear membrane were clearly defined in the absence of nuclear distortion.

Another advantage with Bouin's solution was harder fixation in a relatively short period of time, permitting processing on the short cycle despite only one-hour fixation and enabling the technician to cut excellent thin sections. In most instances, when the half fixed in formalin for periods even up to eight hours was processed on the short cycle, it was extremely soft and either difficult or impossible to cut with the microtome. Acrochordons and cellular nevi posed the most problems in this regard. The sections tended to be too thick and often folded, making histologic interpretation difficult (Fig. 5). Good sections could be obtained only with over-night processing on the long cycle. No significant differences were observed in the tinctorial quality of any of the special stains utilized and there

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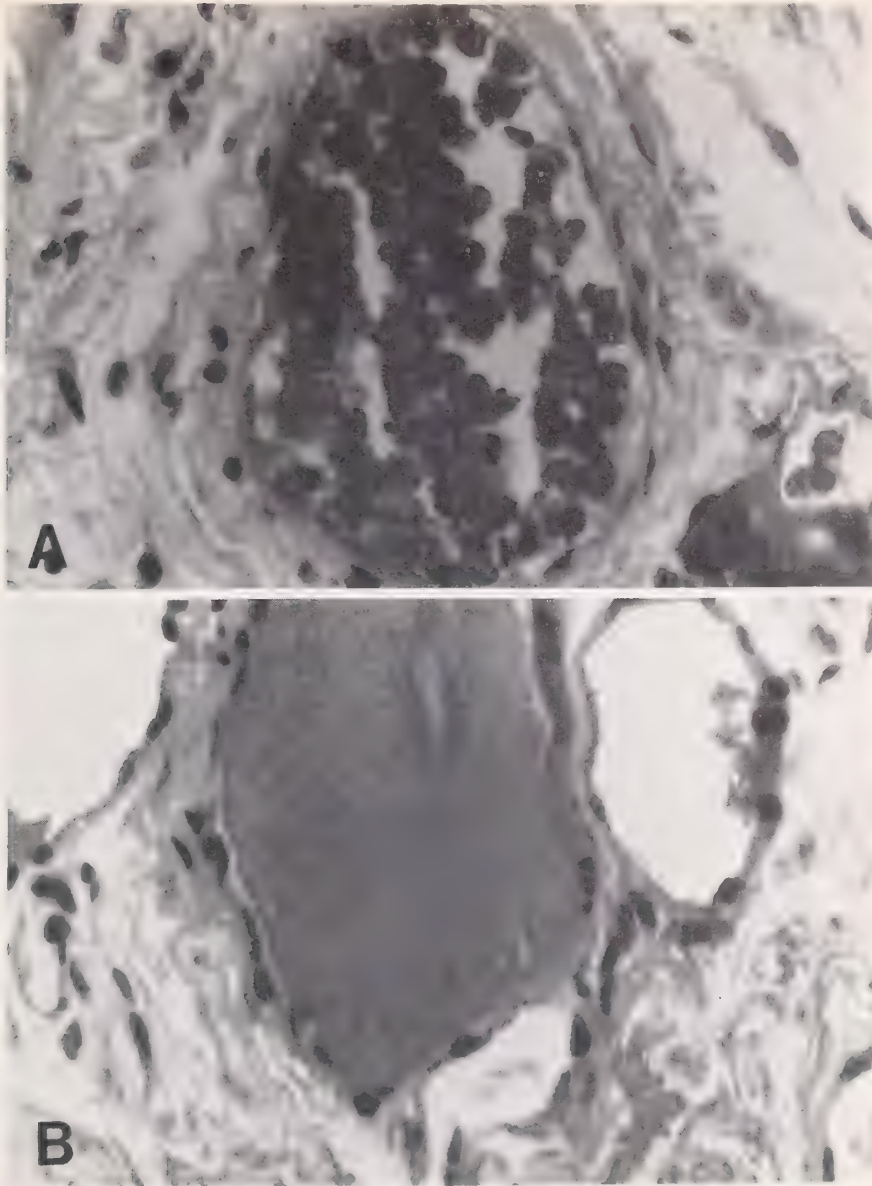


FIG. 1A. Excellent red blood cell preservation with formalin fixation (H & E $\times 400$).

FIG. 1B. Other half of specimen illustrated in Fig. 1A showing hemolysis with Bouin's solution (H & E $\times 400$).

was no appreciable fading or differences in fading during the entire period of study. This was also true of the resectioned slides.

Discussion

From the data obtained, it can be seen that fixation with Bouin's solution has decided advantages over formalin fixation. A good nuclear stain is essential in histopathology. At times, it can decide the difference between a benignity and a malignancy (3). An earlier diagnosis provided by shorter

fixation and processing will permit earlier treatment and, hence, an earlier cure. Bouin's fixation provides both of these very important qualities which are lacking with formalin fixation.

The disadvantages of Bouin's fixation compared to formalin fixation are negligible. These consist of a slightly increased cost of materials, extra care to avoid staining hands and clothing from the picric acid and hemolysis. The latter is probably caused by the low pH (1.4) of Bouin's solution.

My findings contrast sharply with what has been previously stated regarding Bouin's solution (1, 2). In my opinion, it represents a most desirable fixa-

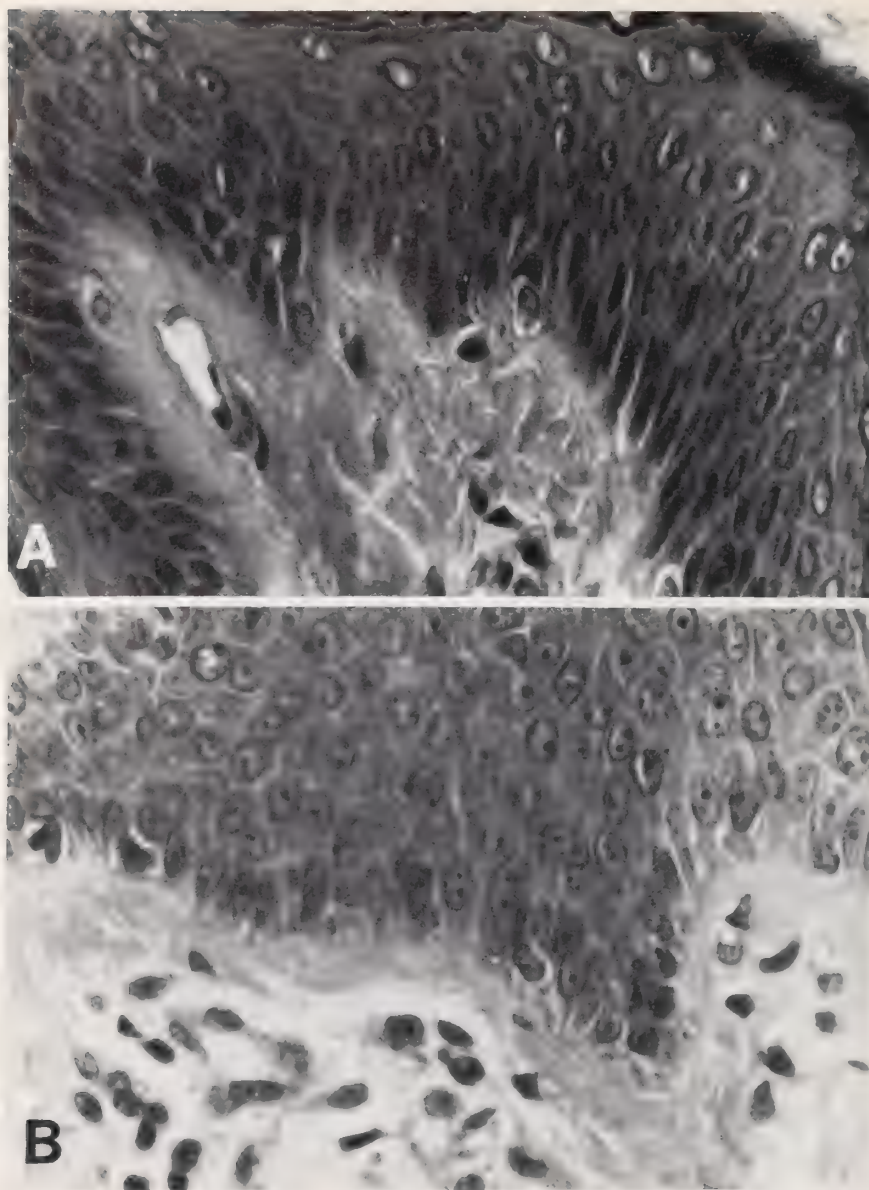


FIG. 2A. High magnification of papilloma showing smudged and pyknotic nuclei with conspicuous linear distortion in basal layer with formalin fixation (H & E $\times 400$).

FIG. 2B. Other half of specimen illustrated in Fig. 2A demonstrating well defined nuclei without distortion with Bouin's fixation (H & E $\times 400$).

tive and not "the least desirable" fixative as stated by the other authors. Contrary to their findings, the specimens are uniformly well fixed allowing even thin sections without displacement of dermal collagen, no formalin crystals are noted, mucins stain just as well as with formalin fixation and no appreciable fading or differences in fading were observed in the routine hematoxylin and eosin stain during the entire period of study. Perhaps the differences between my findings and those of others (1, 2) can be explained on the basis of both more refined vacuum processing with the newer machinery and the meticulous care of the solutions utilized in both processing and routine staining. All solutions must

be exact and uncontaminated, particularly the multiplicity of alcohols used, in order to insure thorough removal of picric acid. Without residual picric acid, the paraffin blocks may be sectioned years later without damage to the tissue.

Other advantages of Bouin's fixation compared to formalin, not associated with this study, include superior preservation of antigenicity of intracellular immunoglobulins (4, 5) and the absence of ice crystal distortion. The artifact of ice crystal distortion, which has been encountered with formalin fixation (6) has never been observed by me despite the fact that thousands of specimens have been exposed to freezing temperatures during the winter months.

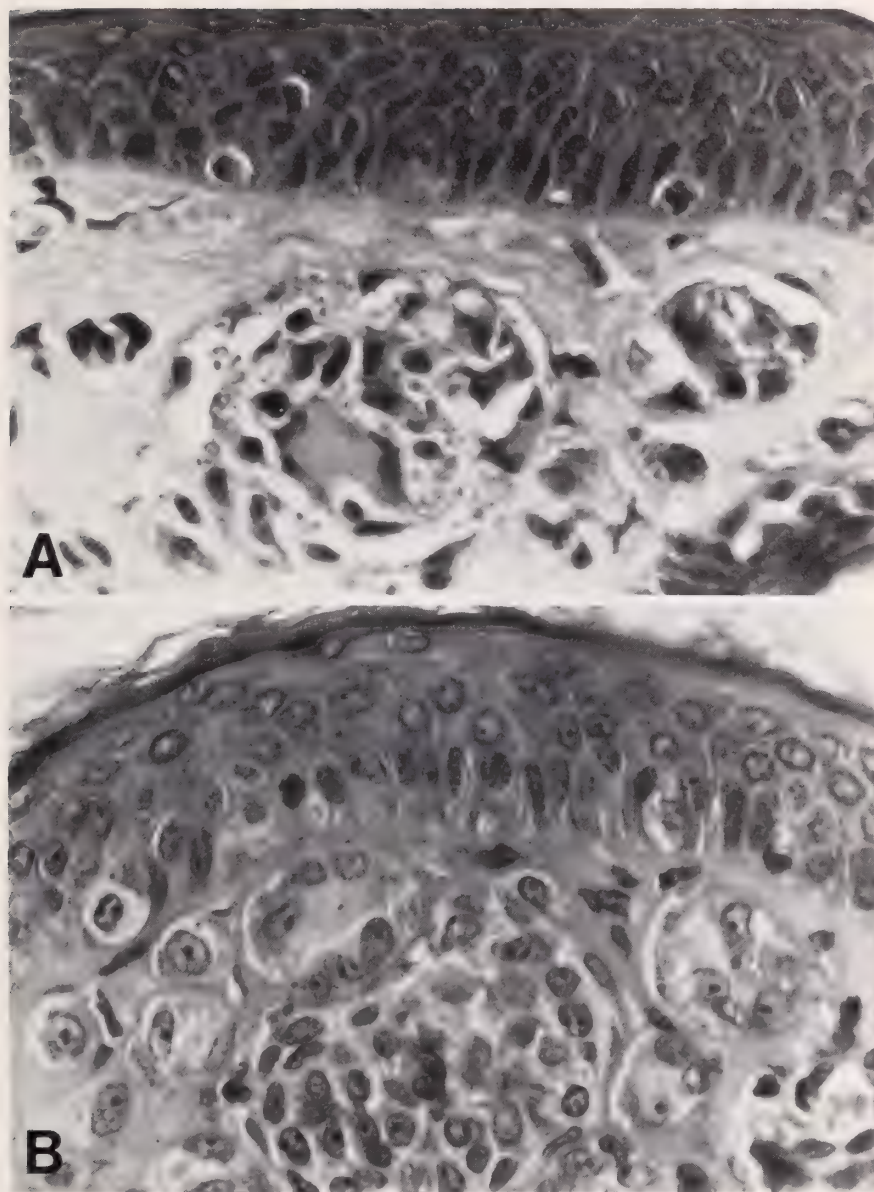


FIG. 3A. Intradermal nevus fixed in formalin showing poor nuclear detail and distortion (H & E $\times 400$).

FIG. 3B. Other half of specimen illustrated in Fig. 3A fixed in Bouin's solution demonstrating excellent nuclear detail without distortion (H & E $\times 400$).

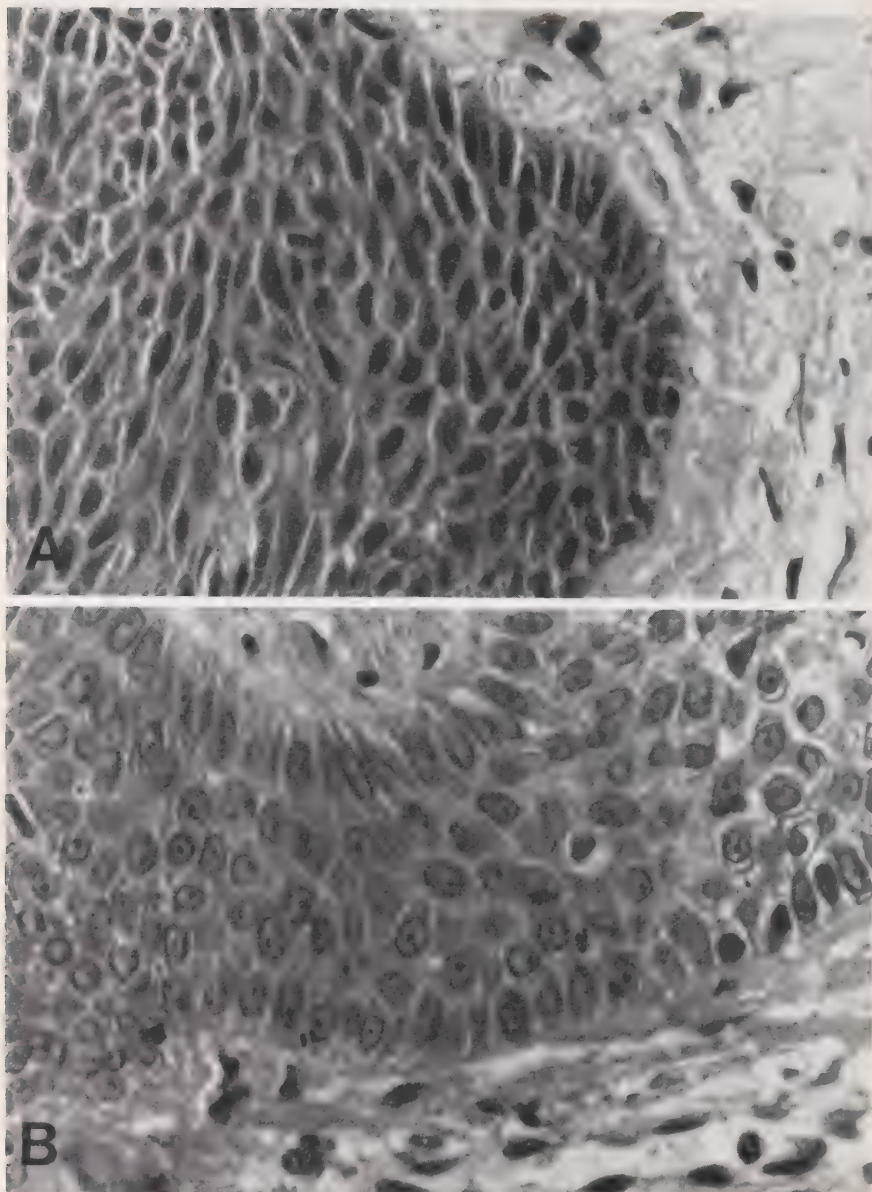


FIG. 4A. Seborrheic keratosis with marked nuclear hyperchromatism and distortion with formalin fixation (H & E $\times 400$).

FIG. 4B. Other half of specimen illustrated in Fig. 4A demonstrating excellent nuclear detail without distortion with Bouin's fixation (H & E $\times 400$).

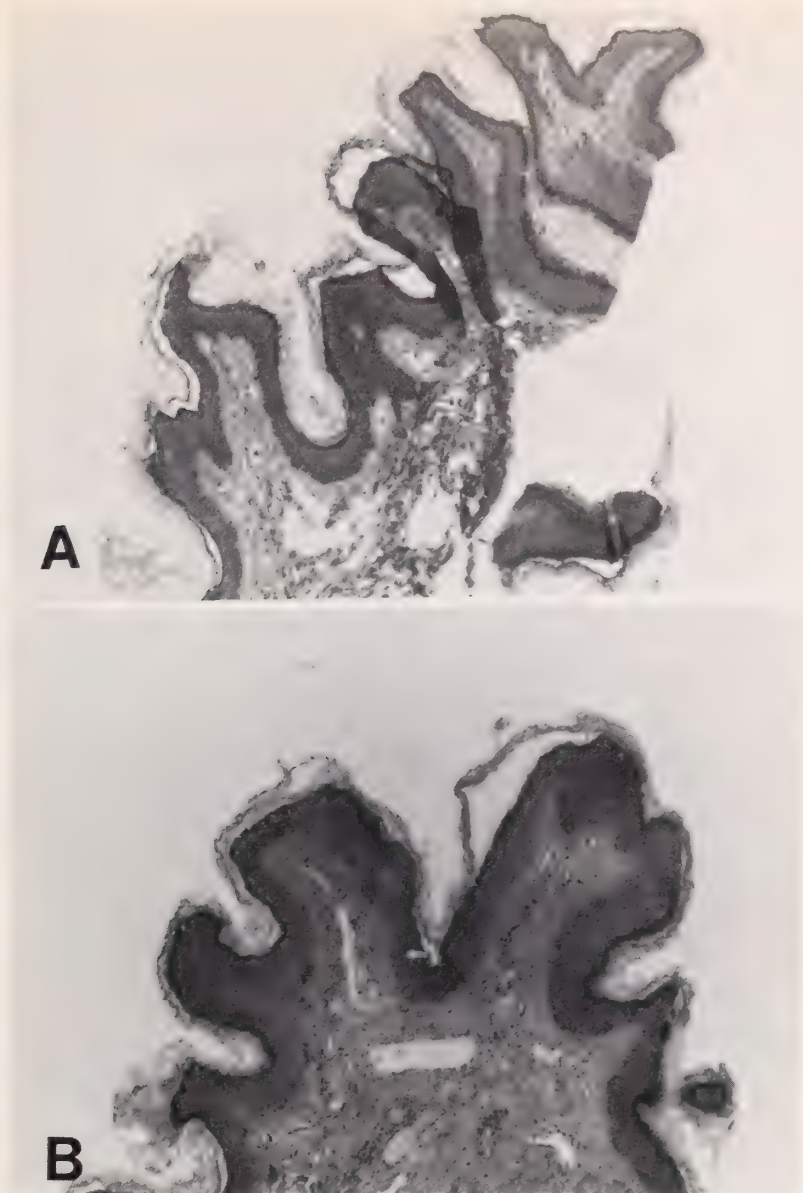


FIG. 5A. Thick, uneven, folded and distorted section of papilloma with formalin fixation (H & E $\times 40$).

FIG. 5B. Other half of specimen illustrated in Fig. 5A showing thin, even nondistorted section with Bouin's fixation (H & E $\times 40$).

Summary

A careful objective comparison of the results of fixation by ten percent neutral buffered formalin and Bouin's solution was performed on 100 consecutive skin biopsies. Bouin's solution affords an earlier diagnosis because of shorter fixation and processing times and provides a far superior nuclear stain than formalin fixation. The disadvantages of Bouin's solution consisted of hemolysis, extra cost and extra care to avoid staining by picric acid, all of which were considered to be relatively significant.

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The Endorectal Pullthrough: Surgical Considerations

TOMAS HEIMANN, M.D.

Total colectomy with mucosal proctectomy and endorectal pullthrough was first described by Ravitch and Sabiston in 1947 (1). This procedure has the decided advantage that it allows the removal of the entire colon and rectal mucosa with preservation of the anal sphincter. Despite this, its use in familial polyposis coli and ulcerative colitis has not gained wide acceptance. The operation has been modified extensively (2-7) but there are still problems caused by the frequent bowel movements in the early postoperative period. By modifying the surgical technique, it has been possible to markedly diminish the postoperative diarrhea and to minimize anastomotic complications.

Surgical Technique

The operation is performed with the patient in the lithotomy position. Following mobilization of the colon, the superior hemorrhoidal artery is identified and ligated. A 2-3 cm longitudinal incision is made in the anterior rectal wall proximal to the peritoneal reflection. The submucosal plane is identified and the surrounding muscular layer is incised circumferentially (Fig. 1). The dissection is continued along this plane to the level of the coccyx. The many small vessels that traverse this plane must be coagulated during the dissection. A large Foley catheter is placed in the rectum and secured with a silk tie (Fig. 2). The terminal ileum is then transected near the ileocecal junction and the colon is separated from the dissected rectal mucosa using the GIA stapling instrument. The procedure is continued from the perineal approach. The mucosa is

everted and transected 1 cm above the dentate line (Fig. 3). The ileum is pulled into the rectal tunnel and sutured to the remaining mucosa using interrupted absorbable sutures (Fig. 4). This anastomosis must be performed without tension and frequently, one or two terminal branches of the ileocolic vessels need to be ligated to achieve this result. A Penrose drain is then brought out through the perineum as described by Shirazi and Soper (Fig. 5) (5). Using the Turnbull technique (8), a loop ileostomy is performed to defunctionalize the anastomosis. Approximately eight weeks later, after the anastomosis is fully healed, the loop ileostomy is closed.

Results

This procedure has been performed in six patients, from 11 to 24 years of age. Four patients had familial polyposis coli and two had ulcerative colitis. The length of postoperative hospitalization was less than 18 days. Complications consisted of one case of deep vein phlebitis which required anticoagulation. One patient had some retraction of the anastomosis which healed without further problems. The others healed primarily, without complications. In all cases, the anastomosis healed within eight weeks and the loop ileostomy was then closed.

This closure was performed by local mobilization of the ileostomy without requiring a laparotomy. Hyperalimentation was not necessary in any of the patients and the frequency of bowel movements was controlled with tincture of opium and methylcellulose. All the patients were discharged within two weeks of admission. They were fully continent and had approximately eight bowel movements per day when sent home. Four patients have been followed over one year. They have four to six formed bowel movements per day and are off all medication.

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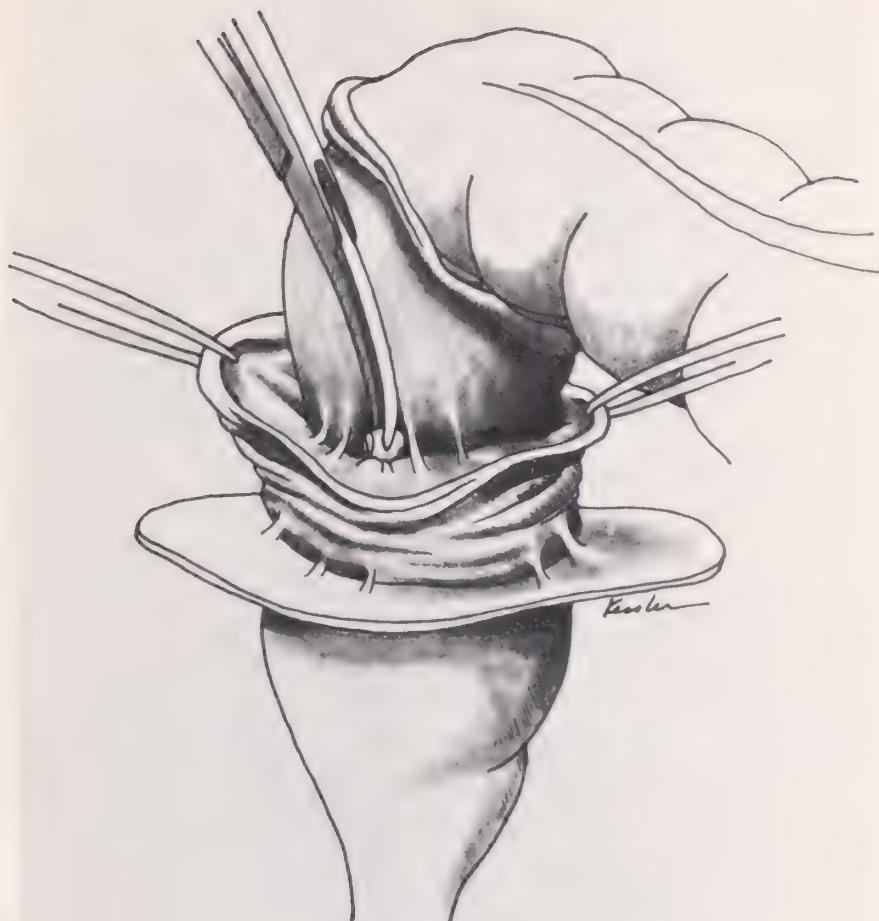


FIG. 1. Shows the initial dissection of the rectal mucosa away from the muscularis.

Discussion

The addition of a temporary loop ileostomy to the endorectal pullthrough has markedly improved the quality of the postoperative convalescence. Although partial anastomotic dehiscence cannot be completely prevented, diversion of the intestinal contents allows the anastomosis to heal without problems. Retraction of the ileum is a consequence of tension on the anastomosis and can be prevented by adequate mobilization.

The dissection of the rectal mucosa proceeds very easily in patients with familial polyposis coli. The mucosal tube is removed in one piece, without

leaving any islands of tissue behind. In ulcerative colitis patients, this dissection is somewhat more difficult and the procedure should only be performed in those with minimal rectal disease.

This operation provides an alternative to total colectomy and ileostomy in the management of familial polyposis coli and selected cases of ulcerative colitis. The addition of the loop ileostomy which has also been advocated by Martin et al (9), prevents postoperative diarrhea and defunctionalizes the anastomosis until it is fully healed. Once the ileostomy is closed, the number of bowel movements is easily controlled and the patients are fully continent.

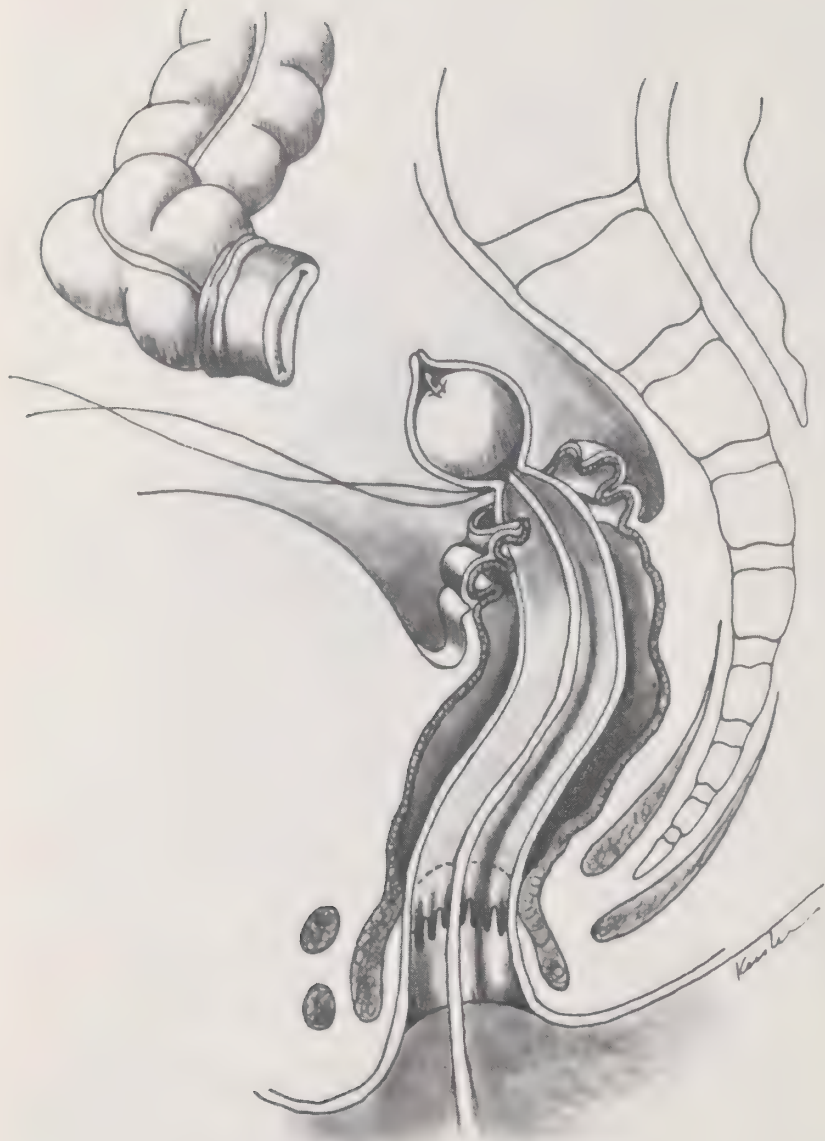


FIG. 2. The mucosa has been separated to just above the dentate line. A large Foley catheter is inserted and secured to the rectal mucosa.

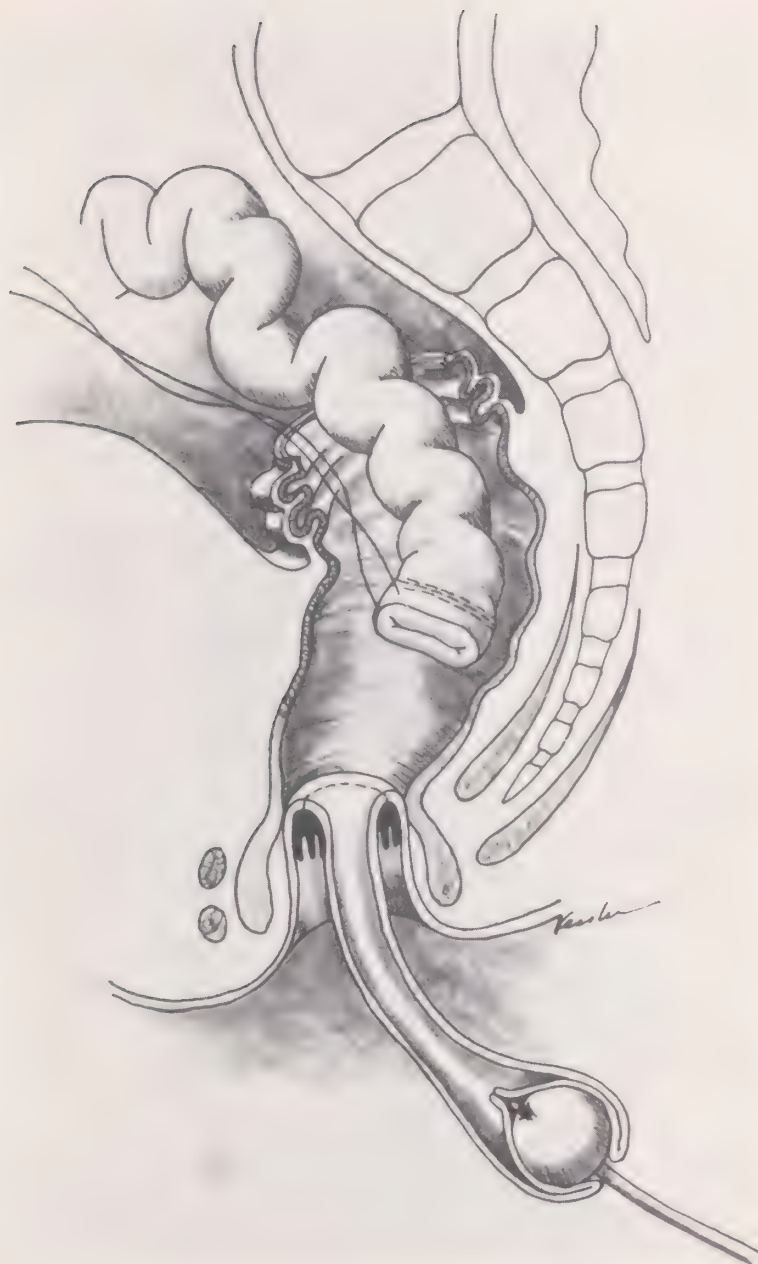


FIG. 3. The mucosa is everted and the terminal ileum is brought through the rectal tunnel.



FIG. 4. The rectal mucosa is excised and an end-to-end anastomosis is performed just above the dentate line.

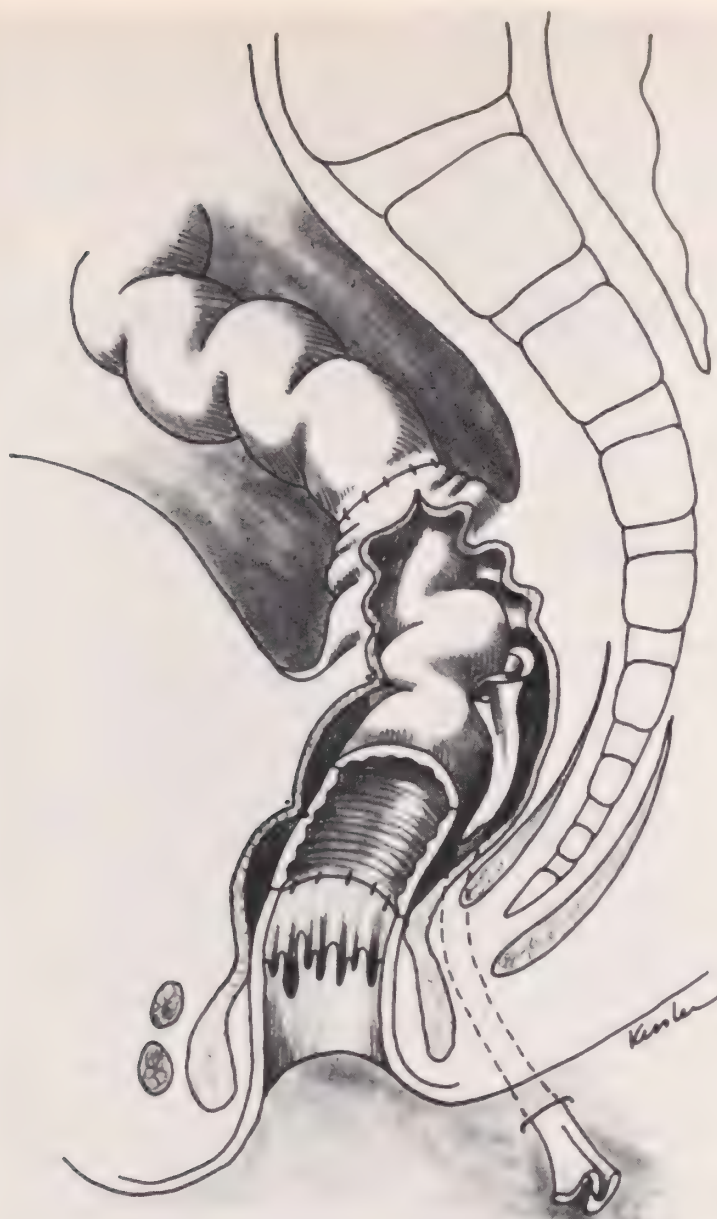


FIG. 5. The area of dissection is drained through the perineum and the rectal muscularis is sutured to the serosa of the ileum.

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Seminars in Reproductive Medicine

Norbert Gleicher, M.D., Editor

Hyperprolactinemic Disorders: An Update

J. VICTOR REYNIK, M.D. AND VIVIAN LEWIS, M.D.

The availability of the radioimmunoassay for prolactin has virtually revolutionized the endocrine aspects of gynecologic practice. Hyperprolactinemia, as the sole or contributing etiologic factor in a variety of menstrual disorders and infertility, can now be found in a significant percentage of patients. Thus, not only the precise initial diagnosis can be reached, but a very specific plan of work up can differentiate between organic and functional causes of the increase in serum prolactin. Finally, currently available therapeutic regimens allow for a specific pharmacologic control of hyperprolactinemic states, resulting in a high percentage of clinical cures. The purpose of this paper is to examine new facts concerning site of production and control mechanism of secretion of human prolactin, review the controversies concerning the plans of work up in hyperprolactinemic states, and suggest our own diagnostic schemes for these patients.

In brief, the main sites of prolactin synthesis and release are anterior pituitary lactotrophs. The normal serum levels during the reproductive years are an average of 2–20 ng/ml, with peaks coinciding with known REM sleep episodes, and with some circadian periodicity. Those variations in levels are lost in patients who manifest hyperprolactinemia.

The levels do not vary greatly during the menstrual cycle and the role of prolactin in regulating the cycle remains unclear in humans. The only well known roles of prolactin are promotion of mam-mogenesis, lactogenesis, and galactoporesis.

However, a new and exciting finding has been reported by Maslar and Riddick¹: the human endometrium in the late luteal phase also appears to be a site of prolactin production. In this study, endometrial tissue was obtained at different points in the menstrual cycle and incubated in Gey's medium and simultaneously dated histologically. No prolactin was found, before or after incubation, in proliferative or early secretory endometrium; but late secretory phase tissue did contain prolactin; which increased in amount after incubation. This increase could be prevented by the addition of protein synthesis inhibitor, cycloheximide, to the medium. The ability to produce prolactin apparently corresponds to onset of histologic decidualization, suggesting a possible role of prolactin in pregnancy implantation.

It has been well demonstrated that human decid-ual tissue obtained from pregnant uteri is capable of prolactin production. Decidual tissue obtained at various gestations (therapeutic abortions, ectopic pregnancies, and term deliveries) has been shown to produce prolactin in vitro, indistinguishable from pituitary prolactin by criteria of immunoprecipitation, gel chromatography, and membrane receptor binding techniques. Placental tissue and fetal membranes, separated from decidua, did not demonstrate this production (2–5). This production was not affected by bromocryptine, dopamine, TRH or previous hypophysectomy (2, 6), but was inhibited

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by protein synthesis inhibitors. This implies that decidua is the source of prolactin present in such high concentrations in amniotic fluid.

The role of prolactin in amniotic fluid is not clearly understood, but it may play some role in osmolar regulation. Animals and human studies have shown prolactin to be a potent modulator of renal excretion of sodium, water, and potassium. (7, 8) The same effect has been demonstrated *in vitro*, using guinea pig amniotic membrane (9). Additionally, in humans, prolactin probably acts to help protect the fetus from rapid gains or losses of fluid secondary to changes occurring in amniotic fluid (10).

Yuen, et al (11) postulated that prolactin may play a role in controlling HCG and estrogen secretion in the fetal-placental unit. It is well known that prolactin affects pituitary gonadotropin release as well as secretion of steroids by the ovaries, testes, and adrenals. The effects on gonadotropin secretion appears to be of direct suppressive type, while in steroids producing tissues, the effect seems variable. Increasing prolactin suppresses steroidogenesis in ovaries and testes, while in the adrenal gland, it appears to enhance androgen production. During pregnancy, the concentrations of HCG appear nearly the opposite to those of prolactin in the maternal compartment. Fetal plasma prolactin rises as the pregnancy progresses. It has been proposed that prolactin, by interacting with estrogens and ACTH, maintains fetal adrenal growth. In the study quoted, the findings were that *in vivo* and particularly during late pregnancy, long-term suppression of prolactin using bromocryptine was associated with enhanced ACTH, estradiol, and estrone concentrations without alteration in progesterone level. These effects appear dependent on diminished prolactin concentration rather than the dopaminergic effect of bromocryptine. These studies open a new field for investigations.

Let's briefly review the control of pituitary production of prolactin in normal women. The prolactin inhibiting factor appears to be represented mainly by dopamine. Dopamine inhibits pituitary prolactin synthesis and release when infused via the portal vein system. Dopamine appears secreted directly into the portal circulation. The release of prolactin appears affected by thyrotropin releasing hormone which, in humans, plays a dual role. Finally, there is an autoregulation of prolactin by a short-loop feedback. By this route, prolactin stimulates dopamine turn over in the tuberoinfundibular neurons and thus increases PIF activity. This autoregulatory mechanism affects also pituitary re-

serve of FSH and LH. Based on dynamic testing, it has been shown that in the presence of increased prolactin, pituitary response to gonadotropin releasing hormone infusion indicates normal synthesis but inhibition of release of both gonadotropins. In clinical testing, hyperprolactinemia is associated with decreased baseline gonadotropin levels and loss of their pulsatile pattern of release.

A recent study (12) suggests a role for catechol estrogens in regulation of prolactin. The metabolism of estrogens in the human proceeds principally by two separate and largely competitive hydroxylation pathways. Hydroxylation at 16- α in the terminal 5-carbon D ring results in formation of estradiol, while hydroxylation in the aromatic A ring results in 2-hydroxyestrone. These dual metabolic routes of estradiol acquired particular significance in light of the divergent biological properties. Estradiol seems to have a potent uterotrophic role equivalent to estradiol, while 2-hydroxyestrone is virtually devoid of such activity. It does, however, exhibit a central action by affecting the release of gonadotropins. Most importantly, there is a substantial *in situ* formation of 2-hydroxyestrogens in the central nervous system. In the study quoted, the findings were that infusion of 2-hydroxyestradiol in estrogen-primed postmenopausal women caused rapid and total suppression of serum prolactin. This represents totally new findings. Recently, 2-hydroxyestrogens have been reported to compete for dopamine receptors in the anterior pituitary and their possible role as dopamine antagonist has been suggested to account for the estrogen induced prolactin release. From the study quoted, however, it appears that in the human, catechol estrogens act as either dopamine antagonist or estrogen antagonist and inhibit prolactin release. Other evidence is now also available linking 2-hydroxylation of estrogens in the brain to prolactin release inhibition. Narcotic agonists which induce prolactin release, inhibit estradiol 2-hydroxylates in the brain while the opiate antagonist, naloxone, which inhibits prolactin release, is an effective inducer of the hydroxylase enzyme. The study suggests the possible new therapeutic implications for 2-hydroxylated estrogens. Another possible mechanism in regulation of prolactin is represented by endorphins.

Recent data indicates that opiated substances inhibit LH release and augment prolactin secretion by the pituitary in rodents, monkeys, and in normal adult males (13-16). These effects can be competitively inhibited by treatment with the opiate receptor antagonist, naloxone (13, 16, 17). While some reports suggest that these substances have a

direct effect on the pituitary gland, it seems more likely that opiate receptors and endorphins modify prolactin secretion by inhibiting dopamine impulse flow from hypothalamus to pituitary, thereby decreasing prolactin secretion (18-22).

Recently (23), evidence was presented for an increased opiate inhibition of LH secretion in patients with hyperprolactinemia due to the pituitary microadenoma. In normal women, naloxone infusion induced no significant change in the levels of FSH, LH and prolactin. In patients with pituitary adenoma, a significant increment in circulating LH occurred, in the form of an amplified pulsatile pattern of release.

Clinical Considerations

Increased prolactin levels may induce a variety of menstrual disturbances:

1. One-third of the patients with secondary amenorrhea manifest hyperprolactinemia. Of these, only 30%-40% have clinically detectable galactorrhea.
2. It appears that 3% of women with primary amenorrhea have increasing prolactin (24).
3. In women with anovulation, there is approximately 80% incidence of increased serum prolactin (25).

In patients with hyperprolactinemia, many attempts have been made to differentiate by dynamic testing the organic and functional causes of the disorder. Thus, stimulatory and inhibitory studies can be performed in such patients.

Stimulation

1. Chlorpromazine test: 25-50 ml of chlorpromazine administered intramuscularly and prolactin level assessed at $\times 0$, 20, 30, and 60. In the normal result, there is a doubling of prolactin level from the baseline. Chlorpromazine stimulates prolactin by suppressing the hypothalamic secretion of PIF.
2. TRH test—500 mcg administered IV with levels of prolactin checked at $\times 0$, 20, 30, 60, and 120 minutes. Again, as a normal response, doubling of prolactin levels is observed. TRH acts directly on the pituitary gland to release prolactin.

Inhibition

1. L-Dopa and bromocryptine test: administration of these drugs results in effective suppression of prolactin levels.

Interpretation

1. Response to chlorpromazine and TRH suggests integrity of both hypothalamus and pituitary.
2. Response to TRH but not to chlorpromazine suggests a normally functioning pituitary gland with a disturbance at the level of the hypothalamus.
3. Failure to respond to both CPZ and TRH indicates disease of the pituitary gland.

However, from the available information, it appears that in the diagnostic process the baseline single prolactin value is of greatest importance. Of all tests, TRH stimulation is the only one offering some hope of being discriminatory for pituitary adenoma. Blunted responses are more likely to occur in the presence of a tumor. Failure to respond to TRH suggest that the tumor cells are already producing at the maximal capacity although the receptor sites for TRH are altered in tumor cells. Because of variable results of such studies, the tests alone cannot permit the diagnosis before the appearance of x-ray studies. Let me reiterate, that the single prolactin value and radiologic study of the sella turcica region are at present the main diagnostic tools. However, even in this area, a new controversy is present. In a study in 1978 (26), the following statements were made about the incidence of abnormal sella turcica architecture in patients with galactorrhea:

1. In presence normal menses and galactorrhea, 6%.
2. In normoestrogenic amenorrhea, 227%.
3. In hypoestrogenic amenorrhea, 67%.

Until recently, sella turcica hypocycolidal polytomography has been recommended as the only reliable method of evaluation of patients with galactorrhea. The amount of radiation exposure to the lens of the eye with this technique is increased as compared with plain A-P and lateral views of the sella; it is 4 $\times$ greater when eye shields are used and 20 $\times$ without eye shielding. Thus, the same group reported a new study in 1979 (27) with the following conclusions about patients with galactorrhea:

1. In a group of patients with normal prolactin, tomography did not detect abnormalities undemonstrated by plain radiography. There were no abnormal tomograms in 80 patients who had normal cone views and normal prolactin levels.
2. In all patients having abnormal AP and lateral films regardless of prolactin levels, tomography did not add any further significant radiographic information. Instead, in these patients,

CAT scan, visual field determination, and possible pneumoencephalography must be obtained since neither plain nor tomographic radiography can detect suprasellar extension of tumor.

3. In contrast to findings in these two groups of patients, no correlation was found between plain films and tomograms in patients with hyperprolactinemia and normal AP and lateral films. Because 7 of 87 patients were found to have abnormal tomograms consistent with adenoma after showing negative plain study, it is necessary only in these patients to perform hypocyctoidal tomograms.

These conclusions are in concept with editorial comments in OB/GYN Survey (28). Reviewing the report of the investigation of over 450 patients, it was noted that correlation exists between the levels of hyperprolactinemia and abnormalities on x-rays of the sella turcica. It was then recommended that tomograms be performed in all patients showing levels of prolactin greater than 100 ng, but after initial screening with AP and lateral films.

To add to the confusion, a sharply contrasting study (29) appeared. Here, in 57 patients with abnormal polytomography, only 37 (65%) have elevated prolactin, and only 16 have galactorrhea. Therefore, 35% of patients with abnormal tomography have normal prolactin levels and five of these had tumors confirmed by surgery.

The controversy will persist for some time. I feel obligated to suggest my own personal plan of work up for patients with menstrual irregularities, anovulation and/or galactorrhea:

1. Single prolactin assay. Random samples are sufficient and diagnostic, as variation in the amplitude of the spikes of secretion and the sleep-related increases are abolished in both functional and tumor-related states of increase of prolactin.
2. All patients with galactorrhea and/or hyperprolactinemia must have assessment of TSH levels.
3. Patients with galactorrhea and normal menses as well as normal prolactin level should be screened with AP and lateral sella turcica films only.
4. Patients with hyperprolactinemia should be screened with AP and lateral films:
 - a. If these are abnormal, one should proceed to CAT scan, visual field determination, and neurologic consultation.
 - b. If these are normal, one should then perform tomography.

Conclusion of work-up:

1. With finding of macroadenoma, neurosurgical consultation and recommendation of extirpation of the tumor be made. It has been estimated that approximately 40% cure rate can be expected after extirpation of macroadenoma.
2. With findings of pituitary microadenoma, the physician should plan accordingly to patient's childbearing wishes:
 - a. In absence of childbearing desire, observation, assessment of prolactin level every six months, and periodic assessment of sella turcica architecture thereafter is mandatory.
 - b. In patient's desirous of childbearing: in presence of neurologic or visual symptoms, transsphenoidal excision of microadenoma should be recommended. If the patient is, however, asymptomatic, the evidence is mounting for the utilization of medical therapy to decrease prolactin level and induce ovulation.

Some investigators believe that any pituitary tumor should be removed before pregnancy is considered since the cure rate is high and complication rate low. Nevertheless, complications may occur after surgical procedures and if an operative procedure can be avoided a favorable result obtained with a known surgical method, that method appears preferable (30). This is applicable specifically because the natural history of pituitary microadenoma is so far unknown.

Bromocryptine revolutionized the therapeutic methodology. This drug lowers prolactin effectively, restores menstruation and ovulation in over 80% of patients, resulting in high pregnancy rates. It has been documented (30) that in the great majority of patients, the pregnancies are uneventful. It appeared that the risk of serious irreversible visual impairment is minimal in adequately supervised patients. If and when symptoms appear during pregnancy, they can be managed conservatively and in extreme situations, surgically. In experimental studies, bromocryptine reduces mitotic activity and inhibits estrogen-induced proliferation of the pituitary lactotrophs. It has also been reported that bromocryptine used in large doses can reduce the size of prolactin-secreting tumors.

Taking into consideration risk-benefit ratio, at the present bromocryptine therapy can be given to patients with microadenomas who want childbearing and who understand the need for close supervision during pregnancy.

Patients with idiopathic hyperprolactinemia in absence of detectable organic pathology should also be treated accordingly to their childbearing wishes. Those seeking childbearing should undergo bromocryptine therapy. Those who do not want to reproduce, should be monitored with prolactin levels every six months and periodic reassessment of sella turcica architecture.

Recently, safety and efficacy of bromocryptine has been reviewed (31). Results of collaborative studies involving 226 patients who did not demonstrate detectable pituitary pathology, have been reported. In 187 patients evaluated for efficacy, 80% experienced reinitiation of menses, or pregnancy without having menses first. The average duration of therapy was 5.7 weeks. There was a significant reduction of galactorrhea in 76% of patients after 6.4 weeks of therapy, and complete suppression in 58% of patients after 12 weeks. Maximum reduction in serum prolactin level occurred during the first four weeks of therapy. There was a strong correlation between the reduction in prolactin level and clinical improvement. Sixty-eight percent of patients experienced mild and transient adverse reaction, consisting mainly of transient hypotension and nausea. The author has concluded that bromocryptine is not a curative drug, however. Amenorrhea/galactorrhea generally recurs in 70%-80% of patients.

In conclusion, we presented some new data pertaining to prolactin secretion in normal and pathologic states. As you can see, the changes in clinical management occur almost monthly, and many questions still remain unanswered. Specifically, the pathogenesis and natural history of pituitary prolactinomas are still unknown. However, there is already some evidence pointing to the dopaminergic neurons dysfunction as a cause of these lesions. As you noted, since the last presentation, we have changed the recommendation in application of tomography to patients with hyperprolactinemia. Most importantly, our therapeutic approach to patients with microadenomas has changed from surgically to medically oriented.

We hope we stimulated thinking and affected somewhat your clinical attitudes towards this commonly encountered problem in Reproductive Endocrinology.

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Ophthalmologic Notes

Keith M. Zinn, M.D., Editor

Massive Scleral Necrosis from a *Pseudomonas* Infection Following Scleral Buckling and Pars Plana Vitrectomy Surgery

KEITH M. ZINN, M.D. AND ANDREW P. FERRY, M.D.*

The incidence of ocular infections occurring after scleral buckling surgery for repair of retinal detachment has been reported as varying from 3% to 10% (1-8) and represents a serious complication that could result in loss of the eye. Once the existence of an infected implant is established, it is usually mandatory to plan for the removal of the scleral buckle as soon as possible and to begin systemic and topical antibiotic therapy immediately. After the implant has been removed, the redetachment rate may approach 33% (7). Of all the organisms cultured from eyes with an infected scleral buckle, *Staphylococcus aureus* and *Staphylococcus epidermidis* appear to be the most prevalent organisms (7). Rarely has *Pseudomonas aeruginosa* been isolated as the offending agent (9). The following case is presented to illustrate the marked scleral necrosis associated with a *Pseudomonas aeruginosa* infection of the globe in a patient who had recent pars plana vitrectomy and scleral buckling surgery (explant).

Case Report

G.T. is a 56-year-old hypertensive Negro woman who in June 1974 had bilateral vitreous hemor-

rhages that followed a "retinal vasculitis," according to her ophthalmologist. There was also a history of pulmonary tuberculosis which was treated with triple therapy in 1969 and had been quiescent up to the time of onset of the vitreous hemorrhages. The patient had been treated with topical atropine and Decadron for her bilateral uveitis.

The ocular findings in June 1974 were as follows: Visual acuity, with correction, was hand motion at two feet in the right eye and counting fingers at six feet in the left eye. There was a full range of motion of either globe in all fields of gaze and the eyes were straight in the primary position of gaze. Function of cranial nerves II, III, IV, V (ophthalmic division), VI, and VII were intact bilaterally except for some constriction of the visual fields of both eyes on confrontation testing. The ocular adnexa, and lacrimal gland apparatus were within normal limits bilaterally, as were the conjunctivae, corneae and sclerae. The intraocular pressures were normal (17 mm Hg in the right eye and 19 mm Hg in the left eye) by applanation tonometry. Slit lamp examination of the anterior chambers revealed them to be clear and of moderate depth, with Grade III open anterior chamber angles with some peripheral anterior synechiae at the 1:00, 4:00 and 11:00 o'clock meridians in the left eye. Both pupils were 6 mm in diameter and fixed, due to previously instilled atropine eye drops. The lenses had 1+ nuclear sclerosis and the right lens had an anterior embryonal (suture) cataract, whereas the left lens had posterior

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subcapsular cataract opacities and vacuoles along the visual axis. The vitreous cavities were filled with dense collections of extravasated blood which precluded any view of the posterior segment. A and B scan ultrasonography confirmed the presence of bilateral dense vitreous hemorrhages, but was negative for a retinal detachment or an occult ocular tumor in either eye.

The vitreous hemorrhages persisted for several months and the patient went to Florida, where she underwent bilateral pars plana vitrectomy surgery and scleral buckling surgery in the right eye (Fig. 1) for repair of retinal detachment.

Several days postoperatively, she developed a purulent discharge in the right eye which was accompanied by severe ocular pain. The patient returned to the New York area and was seen two weeks postoperatively. Her best corrected vision was light perception, without projection, in either eye. There was rubeosis iridis (neovascularization of the iris) in the left eye and the intraocular pressures were 11 mm Hg (normal) in the right eye and 60 mm Hg (markedly elevated) in the left eye by applanation tonometry.

A culture of the discharge material from the right conjunctival fornix grew out *Pseudomonas aeruginosa*. The patient was treated with intravenous gentamycin and with topical gentamycin and atropine for 48 hours, and was then brought to the operating theater where the encircling 360° silicone sponge was removed. The silicone sponge was cultured and grew out *Pseudomonas aeruginosa*. All visible scleral buckle sutures were also removed and the globe and retrobulbar spaces were irrigated with copious amounts of gentamycin ophthalmic solution. The sclera was severely necrotic and a tissue

specimen was processed for histopathologic examination. Postoperatively, the patient was continued on intravenous and topical gentamycin for ten days. The systemic medication was then discontinued after several negative eye cultures. The eye healed, but a retinal detachment remained with vision at the light perception level.

Histopathologic examination of the scleral specimen that was obtained at the time of removing the silicone sponge disclosed a highly necrotic piece of collagenous tissue (Fig. 2). The scleral tissue was densely infiltrated with inflammatory cells and proliferating blood vessels. Most of the inflammatory cells were neutrophils and plasma cells. Many lymphocytes and macrophages were present in areas of suppurative necrosis. The histopathologic picture was one of suppurative necrosis of collagenous tissue (sclera) as well as a granuloma pyogenicum. Bacteria were also noted in the sections.

Discussion

An infected buckle is an ocular emergency because it has the very real potential for going on to an endophthalmitis with loss of vision and, in some cases, loss of the eye itself. Recognizing such an infection may be difficult, especially in the immediate postoperative period when inflammation and ocular pain from the surgical procedure are routinely present. However, severe pain with a sharp loss of vision (compared to the preoperative level) when associated with a persistent ocular discharge should alert the clinician to the possibility of an infected scleral buckle. At this point a culture should be taken.

Once infection has been recognized, an attempt should be made to save the buckle by employing intensive systemic and local antibiotic therapy. This is often unsuccessful and the implant or explant, together with the accompanying sutures, must be removed. If there are signs of intraocular involvement (for example, endophthalmitis), it is imperative that all scleral buckling and suture material be removed as soon as possible.

In the case presented in this paper, the infection arose in the immediate postoperative period and intensive systemic and local gentamycin therapy, coupled with surgical removal of the buckle and its sutures, was responsible for saving the globe from a virulent *Pseudomonas aeruginosa* organism. Unfortunately, the retina was detached before the buckle was removed and the patient was left with only light perception vision.

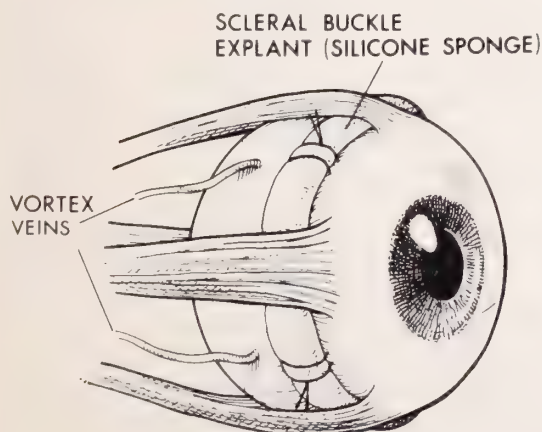


FIG. 1. A 360° encircling scleral sponge used to treat a retinal detachment. Scleral sutures anchor silicone sponge to outer wall of the globe.



FIG. 2. Scleral tissue taken at the time of removing the silicone sponge (Buckle): There is prominent suppurative necrosis of the collagenous connective tissue. A light to moderate infiltrate of neutrophils is present. Hematoxylin and eosin; magnification $\times 120$

Pseudomonas aeruginosa is known to liberate a variety of proteolytic enzymes one of which is collagenase which can degrade collagen (10, 11). The highly necrotic sclera in our case resulted mostly from the release of these proteolytic enzymes, including collagenase. This latter enzyme has the potential of liquifying collagen. More specifically, collagenase will cleave the collagen molecule at one locus through all three polypeptide chains and, once this happens, the particular cleaved collagen molecule falls apart and becomes susceptible to multiple bacterial and tissue proteases (12). Collagenase has been detected on the surface of corneal collagen fibrils, but has not been demonstrated in scleral collagen or in any other connective tissue (12).

Dissolution of collagen is often seen in *Pseudomonas* keratitis, in which actual perforation of the cornea can take place within 24–48 hours after the onset of the infection. This event is believed to be caused, in great part, by the degradative action of the bacterial collagenase. However, the intracellular granules of polymorphonuclear leukocytes contain collagenases and these may have an additive effect in inflammatory states.

Romem et al (9) reported a case of *Pseudomonas aeruginosa* ocular infection developing much later in the postoperative course following scleral buckling surgery. In their case, the infection began about four years after a double cerclage operation was

performed to repair a retinal detachment in that eye. Some authors have found late infections to be more common than early postoperative infections (9). However, Ulrich and Burton (7) reported in their series of 878 retinal detachment operations 37 eyes (4%) in which there was a postoperative infection. In slightly more than half of these cases, the infection occurred within three months of the retinal detachment surgery.

Most authors would agree that infected scleral buckles are caused by organisms introduced at the time of surgery even though the infection itself may not become clinically evident until many months after the surgery was performed (5). Another piece of evidence that may be compatible with this view is that in infected cases (7), only 23% of preoperative cultures correlated with postoperative cultures.

Clearly, there are a number of factors that contribute to the chances of developing an infected buckle and, for the most part, the more damage done to the sclera during the surgery the greater the likelihood of an infection. More specifically, Criswick and Brockhurst (8) reported an infection rate of 6% in a series of cases in which 360° scleral buckling surgery had to be performed. This was an unfavorable rate when compared to an infection rate of 1.3% reported by McMeel (3) in routine scleral buckles performed at the same hospital and group practice. The roles of diathermy, cryopexy,

suture material, buckling material, and technique (implant or explant) in increasing or decreasing the chances for developing an infected eye postoperatively have been reviewed in the literature (7-9, 13).

Postoperative infections occurring in patients who have had scleral buckling surgery for a retinal detachment present difficult management problems that require immediate evaluation, diagnosis and therapy. This often includes appropriate intensive systemic and topical antibiotic therapy and, in many cases, surgical removal of the implant and suture material to save the eye.

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Student's Corner

Paul Celano, B.S., Editor

Pancreatic Calcification and Pancreatic Carcinoma: The Relationship Reconsidered

ROGER E. SCHULTZ, M.D. AND NEIL J. FINKLER, B.A.

Abstract

The presence or absence of pancreatic calcification has been investigated in 181 patients with pancreatic carcinoma, and in 70 patients with chronic pancreatitis. No calcifications were evident in any of these cases of pancreatic carcinoma. Among cases of chronic pancreatitis, calcifications were found in 18%, with a history of moderate to heavy alcohol consumption recorded in all cases of lithiasis. Six (6) cases of pancreatic carcinoma (5%) had a prior history of pancreatitis, none of which had pancreatic stones. This data does not support the idea that chronic pancreatic lithiasis can cause pancreatic carcinoma. Insufficient follow-up data of cases of chronic pancreatitis precludes a definitive statement in this regard. Etiologic variables and associated medical conditions of this series of patients are also reviewed. The data shows high incidences of diabetes and hysterectomy among patients with pancreatic carcinoma. It is suggested that further studies be done to ascertain the importance of diabetes and gynecologic disease to the development of pancreatic carcinoma.

An association between pancreatic calcification and pancreatic carcinoma was first noted by Opitz in 1901 (6). It has been suggested that the severe irritation caused by calcific deposits within the pancreas favors secondary cancerous formations similar to the manner in which chronic cholelithiasis may lead to carcinoma of the gall bladder (7, 9). The practical implications of the association between lithiasis and carcinoma are clear. If, indeed, pancreatic lithiasis is an antecedent to pancreatic carcinoma, one might then be justified in recommending exploratory laparotomy with possible pancreatectomy in patients with pancreatic calcifications.

Several studies report a higher incidence of pancreatic carcinoma in patients with a prior history of chronic pancreatitis, especially chronic pancreatitis with pancreatic calcifications. In a study of 100

patients with chronic pancreatitis, Pauline-Netto et al (1960), found six cases (25%) of pancreatic carcinoma in 24 patients with chronic calculous pancreatitis, and no cases of pancreatic carcinoma in the remaining 76 patients without calcifications (7). Alcohol was the predominant etiology of lithiasis (71%). Howard and Jordan (1960) reported three cases (9.4%) of pancreatic carcinoma in 32 patients with pancreatic lithiasis (3).

Other studies have not yielded such high incidences. Johnson and Zintel (1963) reported a study of 113 patients with pancreatic carcinoma of whom none had a well documented diagnosis of chronic pancreatitis or pancreatic calcifications (4). Of 64 patients with chronic pancreatitis, none had an associated pancreatic carcinoma. In the same article, Johnson and Zintel report their review of the English literature. Of 653 cases of pancreatic calcification, 24 patients (3.7%) were found to have pancreatic acinar or ductal carcinoma. Gambill (1971) reviewed 255 patients with proven pancreatic and ampullary carcinoma and found that 26 pa-

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tients (10.2%) had significant earlier pancreatitis, of these, one patient (4%) had a ductal calculous (2). Leger (1973) reviewed 20 cases of pancreatic lithiasis and found an associated pancreatic carcinoma in one case (5%) (5). Finally, Stobbe (1970) studied 45 cases of pancreatic lithiasis and noted carcinoma in three cases (6.6%) (9). The pathologic findings and clinical symptoms of these three cases suggested that the carcinoma preceded and may have caused calculous deposition, probably by means of mechanical obstruction with stasis.

It is clear that the alleged association between lithiasis and carcinoma of the pancreas is still in dispute. This paper reviews the incidence of pancreatic calcification in patients with chronic pancreatitis and pancreatic carcinoma at The Mount Sinai Hospital between January 1974 and December 1976. In addition, several environmental variables and predisposing conditions are analyzed and discussed in relation to the etiology of these two diseases.

Materials and Methods

The records of all inpatients at The Mount Sinai Hospital from January 1974 to December 1976, with a discharge diagnosis of pancreatic carcinoma or chronic pancreatitis, were retrieved using the Diagnostic Index (PAS) and the Tumor Registry. A total of 181 cases of pancreatic carcinoma and 70 cases of chronic pancreatitis were reviewed. Cases of carcinoma were confirmed by exploratory lapar-

otomy and biopsy, and/or autopsy. The cases of chronic pancreatitis were confirmed by exploratory laparotomy and biopsy, autopsy, and/or a typical history of recurrent attacks of mild to severe pancreatitis, usually with a secretin test indicative of pancreatic insufficiency. Pancreatic calcifications were confirmed either by roentgenographic, operative, or autopsy findings.

Each patient's sex, race, religion, smoking history, alcohol consumption, and associated medical conditions were recorded, in addition to the observation of pancreatic lithiasis. The patients' profiles of this series are presented in Tables I-IV.

Results

Of the 181 cases of pancreatic carcinoma, there were 58 (32%) in which necessary diagnostic studies that would identify pancreatic calcifications were either not done or missing from the chart. In these cases the presence or absence of calcifications remains in question. Of the remaining 123 cases, there were no cases in which there were documented pancreatic calcifications by any diagnostic method. Six cases of carcinoma (5%) showed evidence from the history of earlier acute or chronic pancreatitis.

Of the 70 cases of chronic pancreatitis, 14 (20%) were lacking essential information to confirm the presence or absence of calcifications. Of the remaining 56 cases, there were ten (18%) with well documented lithiasis. Of these ten cases of lithiasis, five were associated with a history of heavy alcohol

TABLE I
Association of Pancreatic Calcification with Chronic Pancreatitis and Pancreatic Carcinoma

	Number	Calcifications Indeterminate*	Cases under Study	Cases of Documented Calcification	Cases of Chronic Pancreatitis with Subsequent Development of Pancreatic Carcinoma	Pancreatic Carcinoma with Prior History of Pancreatitis
Pancreatic Carcinoma	181	58	123	0	—	6
(% of total)		(32%)	(68%)	(0%)	—	(4.8%)
Chronic Pancreatitis	70	14	56	10	0	—
(% of total)		(20%)	(80%)	(17.8%)	(0%)	—

* Insufficient data in chart to determine presence or absence of calcification.

TABLE II
Profile of Inpatients from January 1974-December 1976 with a Discharge Diagnosis of Chronic Pancreatitis or Pancreatic Carcinoma

	No.	Mean Age (yrs)	Sex		Race				Religion			
			Male	Female	Cauc	Negro	Hisp	Other	Jew	Cath	Prot	Other
Pancreatic Carcinoma	181	67.2	92	89	169	7	4	1	127	36	15	3
(% of total)			(51%)	(49%)	(93.4%)	(3.9%)	(2.2%)	(0.7%)	(70.1%)	(20%)	(8.2%)	(1.6%)
Chronic Pancreatitis	70	46.9	36	34	51	15	4	—	18	28	21	3
(% of total)			(51.4%)	(48.6%)	(72.9%)	(21.5%)	(5.7%)		(25.7%)	(40%)	(30%)	(4.3%)

TABLE III
Etiologic Variables/Associated Medical Conditions

	No.	HX Smoking	HX EtOH	Total	Diabetes-Relation of Onset to Diagnosis				Prior Tumor			Gyn Compl**	
					1 yr Prior	Onset Con-current	Onset Subsequent	Onset Undetermined	Total	Mal*	Ben	Total	Hys.

Pancreatic Carcinoma (% of total) 181 47 (26%) 24 (13.3%) 48 (26.5%) 20 (11.3%) 14 (7.7%) 6 (3.3%) 8 (4.4%) 26 (14.4%) 21 (11.6%) 5 (2.8%) 41 (46.1%) 19 (21.4%)

* Malignant tumors—possibly metastatic—2 colonic Ca, 2 gastric Ca, 1 lung adenocarcinoma, 1 melanoma, 1 basal cell Ca, 3 thyroid Ca, 2 prostatic Ca, 5 breast Ca, 1 cervical Ca, 1 uterine Ca, 2 ovarian Ca.

** Gyn Complications—based on denominators of 89 females with pancreatic carcinoma;—includes hysterectomy (hys), menorrhagia, D&D, uterine fibroids, Ca of uterus, cervix, ovary, breast, and benign breast mass.

consumption, and five were associated with both a history of heavy alcohol consumption and gall bladder disease. No cases of chronic pancreatitis with or without lithiasis, had an associated pancreatic carcinoma.

In the series of patients with chronic pancreatitis, the mean age at diagnosis was 46.9 years. The ratio of males to females was about 1:1. Fifty-one were Caucasian (73%), 15 were Negro (21%), and four were Hispanic (6%). Religious affiliations were as follows: 28 Catholic (40%), 21 Protestant (30%), 18 Jewish (26%), and three undetermined (4%).

Of the patients with pancreatic carcinoma, the mean age at diagnosis was 67.2 years. The male to female ratio was also about 1:1. There were 169 Caucasian (93%), seven Negro (4%), four Hispanic (2%), and one undetermined (1%). There were 127 Jewish (70%), 36 Catholic (20%), 15 Protestant (8%), and three undetermined (2%).

Etiologic considerations of the 70 cases of chronic pancreatitis revealed alcohol alone as the probable causative agent in 25 cases (36%), gall bladder disease alone in 19 (27%), and both alcohol and gall bladder disease in 12 (17%). Two cases (3%) were associated with hyperlipidemia, and one case (1%) with hyperparathyroidism. The remaining 11 cases (16%) had an undetermined etiology.

Among the 181 cases of pancreatic carcinoma, a history of smoking was elicited from 47 patients (26%), and a history of moderate to heavy alcohol consumption in 24 (13%). A total of 48 patients were diabetic (27%), of whom 20 (11%) had developed the diabetes at least one year before the carcinoma, 14 (8%) within the same year, six (3%), at some point after the diagnosis of carcinoma and possible surgical treatment, and eight (4%) with uncertain onset. There were 26 cases (14%) with a history of another tumor. Of these, 21 (12%) were malignant and possibly metastatic. These tumors are described in Table III. Of the 89 female patients, there were 41 reports (46%) of some earlier gynecologic complication, including history of hysterectomy, menorrhagia, D&C, fibroids, cancer of the uterus, cervix, ovary or breast, and benign breast mass. Hysterectomy was, by far, more commonly noted than any other gynecologic disorder. There were 19 of 89 women (21%) who had undergone hysterectomy at some point in their medical history.

Discussion

The incidence of pancreatic calcification in patients with chronic pancreatitis is not rare, occurring in 18% of patients in our series. This figure is

TABLE IV
Etiologic Variables/Associated Medical Conditions

	No.	HX—EtOH Alone	HX—Gall- bladder Dis- ease Alone	HX—Both EtOH & GB Dis.	Hyper-lipi- demia	Hyper-pa- thyroidism	Etiology uncertain	Gyn Compl*	
								Total	Hys.
Chronic Pancreatitis	70	25	19	12	2	1	11	13	5
(% of total)		(35.7%)	(27.1%)	(17.1%)	(2.8%)	(1.4%)	(15.7%)	(38.2%)	(14.7%)

* Gyn Complications—based on denominators of 34 females with chronic pancreatitis;—includes hysterectomy, (hys.), menorrhagia, D&C, uterine fibroids, Ca of uterus, cervix, ovary, breast, and benign breast mass.

less than that reported by Pauline-Netto et al (24%), and Johnson and Zintel (24–38%) (4, 7). A history of moderate to heavy alcohol consumption was obtained from all patients having pancreatic calcification. Of patients without calcification, alcohol was the likely cause in 29%. This data is consistent with earlier studies, and again points to the importance of alcohol in the etiology of pancreatic lithiasis. It is postulated that the epithelial alteration induced by alcohol results in a reduced water and bicarbonate output, which in turn causes intraductal stasis of pancreatic secretions with calculous formation. As an alternative hypothesis, an altered epithelium with increased absorptive capacity would cause inspissation of pancreatic secretions and calcium deposition (9).

Several earlier studies suggested an association between chronic calculous pancreatitis and pancreatic carcinoma (3, 7). The more severe inflammation caused by the presence of calculi may lead to atypical proliferative changes of the ductal epithelium. On the other hand, others argue that calculi, if present, may be the consequence of obstruction by tumor with stasis (4, 9).

In our series of 181 cases of pancreatic carcinoma, it was hoped that there would be evidence of earlier inflammation and/or lithiasis. Only six cases (5%) had well documented clinical proof of earlier pancreatitis. There were no cases of pancreatic calcifications. Thus, among these cases, no evidence was found which would support the idea that chronic calculous pancreatitis would progress to pancreatic carcinoma.

Of 70 cases of chronic pancreatitis, there was not a single case in which pancreatic carcinoma was noted, regardless of the presence or absence of calcifications. Assuming a lengthy latent period in a progression from severe inflammation to carcinoma, prolonged follow-up of these 70 cases would be essential. Failing this information, the combined data of both series of cases are, at best, suggestive that chronic pancreatic lithiasis bears no causal relationship to pancreatic carcinoma. A controlled

prospective study of this question would permit a more definitive statement.

Tables II, III and IV present descriptive findings of our series of patients. Patients with pancreatic carcinoma have a mean age at diagnosis of 67.2 years, with a male to female ratio of 1:1. The reported mean age of diagnosis is 64.4 years, with male predominance (1.5:1) (10, 11). That the male:female ratio is more even in our study probably reflects the small sample size rather than a change from what has been reported.

Jews are predisposed to pancreatic carcinoma (11). Our series show that 70% of the patients were Jewish; however, these data are uncontrolled, and the patient population at The Mount Sinai Hospital consists of proportionately more Jews than at many other centers.

Surveys indicate an approximately two-fold increased risk of pancreatic carcinoma in cigarette smokers (11). Other etiologic variables that are being studied with regard to pancreatic carcinoma include alcohol, occupational exposures, western diet, and viruses. In addition, diabetes, female endocrinopathy and gall bladder disease have been alluded to as possible predisposing conditions (1). Our figures reveal a 27% incidence of diabetes among the cases of carcinoma, of whom 11% had clearly developed the diabetes at least one year prior to the carcinoma. Whether or not the diabetes was a predisposing factor or simply a prodromal sign would require further investigation using matched controls.

Of particular interest was our finding of gynecologic complications in 46% of females with pancreatic carcinoma. Soloway and Sommers reported an increased incidence of ovarian cortical stromal and endometrial hyperplasia, as well as neoplasms of the breast, ovary, and uterus in females with pancreatic carcinoma (8). They have suggested that this association may represent a response of the pancreatic-biliary duct system to excessive gonadotropin stimulation. In our series, a history of hysterectomy was obtained in 19 cases (21%); however,

the reasons for the hysterectomies were usually not recorded. That 21% of the females had a hysterectomy performed seems somewhat high by crude comparison to statistics regarding hysterectomy rates in the Northeast (9). Life table analysis and selection of matched controls for these cases would help clarify this issue.

Summary

1. The incidence of pancreatic calcification was investigated in a series of 181 patients with pancreatic carcinoma, and 70 patients with chronic pancreatitis.
2. No cases of pancreatic carcinoma showed evidence of pancreatic calcification.
3. Pancreatic calcification occurred in 18% of the cases with chronic pancreatitis. No cases of chronic pancreatitis, calculous or acalculous, had an associated carcinoma of the pancreas.
4. A history of moderate to heavy alcohol consumption was documented in all cases of chronic pancreatitis with calcification. In contrast, alcohol was the probable cause in 29% of cases of chronic pancreatitis without calcification.
5. There is no evidence from this study to support the claim that chronic pancreatitis with calcification may presage the development of pancreatic carcinoma. The data, however, is not sufficiently complete to deny the possibility.
6. A descriptive profile of the patients under study is presented. The observations of high incidences of diabetes and hysterectomy among patients with pancreatic carcinoma suggest the need for further studies of these medical complications in reference to the development of pancreatic carcinoma.

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The Incidence of Cancer in Psychiatric Patients

Cancer and the Emotions: A Review

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Abstract

This paper will deal with the general discussion of emotions and cancer. It will be divided into a section on the historical critique of psychological states and malignancies, the incidence of major psychosis and cancer, and the various biochemical hypotheses for the correlation of schizophrenia and neoplasms. This will lead to a realization that depressive states can be shown to precede the clinical appearance of tumors and that the schizophrenic population has a decreased risk of malignancy.

Introduction

The emotional state as a causative factor in cancer has been recognized for many centuries. Many physicians have believed that patients suffering from cancer exhibited certain types of psychological characteristics prior to the discovery of the neoplasm such as hopelessness, helplessness, melancholia and, in general, states of depression. Galen, "Believed that melancholic women were more prone to cancer than those of sanguine temperament (1)." During the 1600's and early 1700's, the humoral theory gained respect with regard to cancer but in the late eighteenth and nineteenth centuries many articles were published concerning the effect of emotions on the physiology of the body. Burrows, in 1767, stated that cancer is caused by "the uneasy passions of the mind with which the patient is strongly affected for a long time, that weaken the circulation of the blood and consequently thicken it (2)."

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It was left to the surgeons who had the most contact with cancer patients to note the specific correlation between cancer and emotions. Walshe wrote:

"Much has been written on the influence of mental misery, sudden reverses of fortune, and habitual gloominess of temper on the deposition of carcinomatous matter. If systematic writers may be credited, these constitute the most powerful cause of the disease; and although the alleged influence of mental disquietude has never made matter of demonstration, it would be vain to deny that facts of a very convincing character, in respect of the agency of the mind in the production of this disease, are frequently observed. I have myself met witnesses in which the connection appeared so clear and decisive, that to question its reality would have seemed to struggle against reason (3)."

Nunn (4), in the late 1800's, believed that the social state and emotional factors could influence the growth of neoplasms. Amussat wrote, "The influence of grief appears to me to be in a general way the most common cause of cancer (5)." Sir James Paget, a renowned surgeon and pathologist, relied on his countless dealings with the subject to write:

"The cases are frequent in which deep anxiety, deferred hope, and disappointment are quickly followed by the growth or increase of cancer. That we can hardly doubt that mental depression is a weighty addition to the other influences that favour the development of the cancerous constitution (6)."

Parker, also a noted surgeon of the nineteenth century, wrote the following:

"Now, can anxiety be a predisposing cause? Will a long period of care, trouble, and sorrow alone disturb the balance between the nervous and cellular elements, so as to make the latter take on an abnormal, a cancerous development? It is more than probable; but must we on that account reject the probability? Are we justified in rejecting every hypothesis which cannot be placed upon a demonstrated basis? . . . But if we believe mental affliction to be one of the predisposing causes of cancer, we shall advise those who have apparently benign tumors in their breast of the advantage of being cheerful (7)."

Snow, an expert on cancer in his time, expanded on the views of his contemporaries and relying more on his clinical observations wrote the following:

"That with carcinoma of the breast and uterus, failing a history of traumatism, we invariably find certain immediate antecedents, trouble of mind and anxiety are the most constant. Conversely we never see malignant disease of the pathological species in question (except as the result of injury), developed in the mamma of a woman leading a healthy, happy well balanced life (8)."

In the 1900's, researchers began to clearly delineate the psychological factors that they felt preceded the occurrence of cancer. Three major categories of concern were pursued: The correlation of stress to cancer, personality traits and their relationship to cancer and the phenomena of hopelessness and loss as precursors to neoplasms.

Stress and Cancer

Researchers have posited an association between stress and cancer. Many have attempted to measure stress and have concluded that prolonged or excessive stress contributes to the etiology of cancer. Holmes (9) categorized and quantitated the responses of a population to various life events. He endeavored to measure stress and its effects as seen in a particular disease state. Holmes concluded that, "The concept of life change appears to have relevance to the causation of disease." In addition, he observed that the effects of certain events were different for various people and suggested a "threshold of resistance."

Many investigators such as Wolf (10, 11) recognized the association between stress and anxiety and have attempted to measure the concomitants of anxiety. Sachar concluded that the effectiveness of coping mechanisms rather than stress itself led to disease. In measuring endocrine disturbances, Sachar observed that:

"When the defense pattern, whether it be psychotic or neurotic is effective in protecting the individual from emo-

tional distress, endocrine disturbance is minimal . . . Psychological defenses would be regarded as operating to minimize emotional disturbance and its correlated endocrine disturbance and this could be viewed as playing an important role in physiological as well as psychological homeostasis (12)."

Schonfield's study (13), corroborates Sachar's hypothesis. On the basis of MMPI data, he determined that women with benign tumors of the breast had higher measures of stress. The coping mechanisms of these women apparently ameliorated the seriousness of the disease.

Personality Traits

Some researchers have emphasized the personality traits of cancer victims in an attempt to discern an etiology for the disease. Kissen (14) theorized that cancer is associated with those individuals who have difficulty in expressing their emotions. Their personality pattern was characterized by "a restricted outlet for emotional expression." These subjects were found to have a low incidence of acting out behavior as children. Grissom, Weiner and Weiner (15) observed cancer patients were more preoccupied with questions of morality than control groups. These findings suggested to them that cancer patients tended to be emotionally unresponsive.

Bahnsen and Bahnsen studied the ego defense mechanisms of their cancer patients. They discovered a high incidence of repression and denial in coping with cancer. They postulated that denial and repression "block any behavioral or cognitive outlet for disturbing impulses; rather they must cope with drives, not psychologically, but somatically, through internal discharge mechanisms (16)."

Tarlau and Smalheiser (17) in 1951 found that women with malignant tumors of the breast and cervix did indeed have a consistent personality pattern when compared to controls for each group. They concluded that women with oral conflicts developed breast cancer whereas women with genital conflicts developed cancer of the cervix. Tarlau and Smalheiser believed that these personality factors were etiologic in nature rather than the result of cancer.

Abse et al also studied the personality characteristics of cancer patients. They found that differences in the personalities between cancer patients and controls were more significant when the ages of the cancer patients were taken into consideration. Older cancer patients did not differ substantially from their matched controls but the younger age group did. The younger cancer patient,

"smokes more, recalls fewer dreams, and shows greater marital stability than do the control patients... He tends to be overly conscientious in his work attendance, shows a marked constriction in his interpersonal relationships, and reports less adequate or frequent sexual relationships. Finally, he is less active socially and appears to have more problems in the handling of dependency needs (18)."

Goldfarb (19) attempted to classify the personality traits of cancer victims emphasizing such characteristics as unexpressed hostility, pented anger, helplessness, despair, hopelessness, immature sexual outlook and overbearing domination by an authority figure.

In continuation of Bahnson and Bahnson's earlier works, Thomas researched the early family dynamics of cancer patients in a prospective study. These patients were not close to their parents when compared to control groups.

"The low closeness scores of the Major Cancer Group may reflect the presence of unresolved psychological problems in the realm of family relationships, rather than merely represent more candid appraisals of family attitudes by subjects who later developed cancer (20)."

It should be noted, however, that Watson and Schuld in 1977 found that there was no support in their study for a "psychogenic theory of neoplastic growth (21)." They attributed the previous positive findings demonstrated by other authors to the retrospective nature of their studies. However, this criticism is invalidated with regard to the prospective Thomas study.

Hopelessness and Loss

The relationship between hopelessness, depression, and loss to cancer has been studied in detail by various investigators. Based on her review of numerous cases, Evans (22) in 1926, postulated that cancer patients as a group lost an important emotional relationship before the discovery of the malignancy. This loss included death or separation from a significant person or thing, the inability to attain wanted goals or the abrupt change in a harmonious environment. Schmale (23) significantly predicted the presence of cancer based on feelings of hopelessness. He found that many of his cancer patients experienced these feelings of hopelessness for long periods before the clinical appearance of disease. Loss was also contributed to the etiology of any ensuing cancer. Schmale reported that:

"These individuals were unable to defend against reminders of previous loss. These patients showed a high incidence of object loss in early life and/or life-long feelings of unsup-

ported dependant needs and rejection. Uncompleted mourning over past object losses was also common (24)."

The concepts of bereavement, loss and separation leading to disease were also championed by Parkes (25).

LeShan (26) wanted to test the relationship of personality traits and loss to cancer. He found that the loss of a love object, despair and unexpressed hostility were statistically significant with regard to cancer patients compared with a normal control group.

Recently, some authors have disagreed with Schmale and Parkes. In a small study, Niemi (27) found that serious depressive illnesses did not lead to any increase in cancer morbidity. Greenwald et al (28) found that the increased incidence of prostatic cancer in widowers was related to "artifacts of classification" rather than to stress or separation. These studies can also be viewed in light of Sachar's theory of effective coping mechanisms however, and not just the presence of depression or loss.

Schizophrenia and Cancer

The correlation between cancer and schizophrenia is still being debated. Beginning in the early 1900's two groups of opinions developed in response to reports that there were few cancer deaths in the large mental institutions of Europe and the United States. Some investigators felt insanity might be a saving grace with regard to developing neoplasms while others showed evidence of no statistical difference between death rates caused by cancer in a psychiatric population compared to controls. As early as 1909 it was written: "There remains one morbid condition (cancer) which is responsible for an increasing number of deaths in the general community, from which it would almost appear as if the insane enjoyed some immunity (29)." In the early 1930's the same results were observed; the insane had one third less cancer mortalities, but various circumstances questioned the conclusions. For instance, there was a plethora of other disease which claimed the lives of the mental patients and an environment in the institutions which bred infectious disease. Could these patients have been dying from other factors before cancer developed? In the 1930's, Lewis (30) described paranoid schizophrenics as having a greater risk of malignancy. This was one of the first attempts to subdivide the category of schizophrenia in its relationship with cancer. Previously all mental illnesses were discussed together when dealing with cancer mortality rates.

Statistical Evidence?

Scheflen (31) in the 1950's attempted to reconcile the diverse viewpoints concerning mortality rates and the different conclusions which had been drawn. Some said the insane population had a much lower incidence of cancer while others claimed that the mental institution population had a higher incidence. Scheflen reconciled these contrasting viewpoints by explaining them in terms of different statistical methods employed.

"A low incidence will be consistently found by basing the result on proportionate mortality rates; a higher incidence when cancer death rates are employed (32)."

The proportionate mortality rates are only applicable when the death rates are approximately equal. Cancer death rates provided a more accurate evaluation. With these statistical methods in mind, Scheflen concluded that the psychotic patients had a higher frequency of cancer than the control populations. The type of cancer did not differ in its incidence in these two populations except for cancer of the pancreas which was found more in schizophrenics (33). Scheflen specifically sought to categorize the different psychoses with regard to their specific cancer death rates. He stated that the higher incidence of cancer was due to the higher incidence of neoplasms in the senile psychotics and psychotics with cerebral arteriosclerosis. The skewed distribution in the institutions of such groups generated higher cancer death rates among the psychotic patients than the control populations. Paranoid schizophrenics also showed higher cancer death rates. Hebephrenics, catatonics, schizo-affectives and depressive psychoses showed a much lower incidence. Therefore, even the general statement that psychotic patients had a higher incidence of cancer must be examined more carefully. Ehrentheil (34), facing almost the same data and working in the same geographical area and time came to different conclusions. He felt that there was no difference in cancer rates between the control and psychotic population, not even among the paranoid schizophrenics. Writing in the early 1970's Rassidakis et al (35) found that only 4.9% of the mentally ill died from cancer as compared to 15% of the general population. It becomes clear that this subject is not easily reconcilable.

Pathophysiology

Dopamine Levels

Beginning in the late 1950's, the correlation between mental illness and cancer had to be further

examined. Was it the increased dopamine of the untreated schizophrenic or the reduced levels in the patients receiving phenothiazines which led some researchers to conclude that schizophrenics rarely developed cancer. In a study from the late 1970's Modrzewska and Böök (36) found reduced cancer mortality in a group of schizophrenics. There was no mention of whether this group had received phenothiazines or other neuroleptic drugs. Other studies are needed to separate that point of controversy, but these investigators showed evidence of genetically determined psychosis due to inbreeding. This group had low levels of dopamine beta hydroxylase and monoamine oxidase. This deficiency of catecholamine metabolism led to a dopamine excess. Was it this dopamine surplus that caused the lessening of the incidence of cancer in this controlled group in Scandinavia?

Neuroleptics and Cancer

At this point, it is necessary to examine the effects of the neuroleptics used for schizophrenic patients. Will they allow schizophrenics to approach a higher cancer rate? There has been little written on this subject but Schyve, Smithline and Meltzer (37) have shown that neuroleptics can indirectly induce the growth of cancer of the breast. Neuroleptics cause the increase of prolactin levels by decreasing dopamine levels which inhibits the release of prolactin inhibiting factor. The increased prolactin causes a hyperplasia and malignant change in breast tissue.

Error of Metabolism

At this date, the consensus is that schizophrenics do indeed have a lower incidence of cancer. Levi and Waxman maintain that this could be due to an error of metabolism.

"The true schizophrenic patient could have symptoms which are related to a lack of labile methyl groups although the level of their folic acid is within normal limits and their diet might be adequate for methionine. We postulate that the schizophrenic patient is unable to utilize methionine as a methyl donor because of a deficiency of methionine adenosyl transferase (38)."

Certain tumor cells need an excess of methionine groups to sustain growth. Therefore, if schizophrenics cannot generate free methionine groups, then it follows that malignant cells will fail to thrive and schizophrenics will have a reduced incidence of cancer.

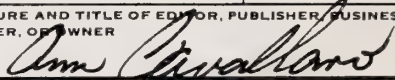
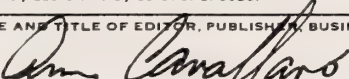
Summary

The question of how emotions and mental states affect cancer is not an easy one to answer. At the present state of knowledge, it is assumed that depression has a direct relationship to cancer and schizophrenia has an inverse relationship. We have tried to summarize the various attempts to answer the question but the definitive answer still eludes investigators.

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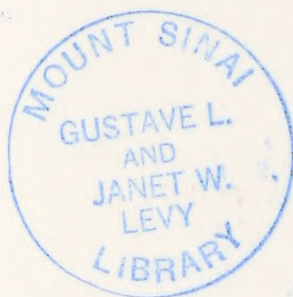
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